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(FILE 'HOME' ENTERED AT 16:13:05 ON 06 SEP 2006)

FILE 'CAPLUS, MEDLINE' ENTERED AT 16:13:24 ON 06 SEP 2006

| | | | | | | | | | |
|-----|-------|---|-----------------------------------|-----|----------------|----------------|---------------------|----------------|----------|
| L1 | 3 | S | ACARBOSE | (P) | ALCOHOL? | (P) | CHROMATOGRAPHY | (P) | ENZYME? |
| L2 | 5 | S | ACARBOSE | (P) | ALCOHOL? | (P) | ENZYME? | | |
| L3 | 2 | S | L2 NOT L1 | | | | | | |
| L4 | 3 | S | ACARBOSE | (P) | ALCOHOL? | (P) | CHROMATOGRAPHY | | |
| L5 | 1 | S | ACARBOSE | (P) | ETHANOL? | (P) | CHROMATOGRAPHY | | |
| L6 | 5 | S | ACARBOSE | (P) | ETHANOL? | (P) | ENZYME? | | |
| L7 | 3 | S | ACARBOSE | (P) | PURIFICATION | (P) | ENZYME | (P) | AFFINITY |
| L8 | 3 | S | ACARBOSE | (P) | PURIFICATION | (P) | FERMENTATION BROTH? | | |
| L9 | 14 | S | ACARBOSE | (P) | PURIFICATION | (P) | CHROMATOGRAPHY | | |
| L10 | 57 | S | ACARBOSE | (P) | ENZYME? | (P) | AFFINITY | | |
| L11 | 1 | S | ACARBOSE | (P) | ENZYME? | AFFINITY | CHROMATOGRAPHY | | |
| L12 | 25 | S | ACARBOSE | (P) | AFFINITY | CHROMATOGRAPHY | | | |
| L13 | 26084 | S | L12 NOT L9 | | | | | | |
| L14 | 17 | S | L12 NOT L9 | | | | | | |
| L15 | 0 | S | ACARBOSE | (P) | ETHYL ALCOHOL? | CHROMATOGRAPHY | | | |
| L16 | 0 | S | ACARBOSE | (P) | ETHYL ALCOHOL? | (P) | CHROMATOGRAPHY | | |
| L17 | 0 | S | ACARBOSE | (P) | ETHYL ALCOHOL? | (P) | PURE | | |
| L18 | 0 | S | ACARBOSE | (P) | ETHYL ALCOHOL? | (P) | PURIFY | | |
| L19 | 0 | S | ACARBOSE | (P) | ETHYL ALCOHOL? | (P) | PURIFICATION | | |
| L20 | 3 | S | ACARBOSE | (P) | ETHANOL? | (P) | PURIFICATION | | |
| L21 | 47 | S | ACARBOSE | (P) | ENZYME | (P) | CHROMATOGRAPHY | | |
| L22 | 1 | S | ACARBOSE | (P) | IMMOBIL? | ENZYME | (P) | CHROMATOGRAPHY | |
| L23 | 0 | S | ACARBOSE | (P) | ENZYME | CHROMATOGRAPHY | | | |
| L24 | 10 | S | ACARBOSE | (P) | ENZYME | (P) | SUPPORT | | |
| L25 | 112 | S | ACARBOSE | (P) | ?GLUCOAMYLASE | | | | |
| L26 | 18 | S | L25 AND PURIFICATION? | | | | | | |
| L27 | 1 | S | L25 AND PURE | | | | | | |
| L28 | 8 | S | L25 AND COLUMN | | | | | | |
| L29 | 20 | S | L25 AND CHROMATOGRAPHY | | | | | | |
| L30 | 14 | S | ACARBOSE/TI AND CHROMATOGRAPH?/TI | | | | | | |
| L31 | 0 | S | ACARBOSE | (P) | AMMONIA | (P) | CHRMATOGRAPHY | | |
| L32 | 3 | S | ACARBOSE | (P) | ETHANOL | (P) | PURIFICATION | | |
| L33 | 4 | S | ACARBOSE | (P) | ETHANOL | (P) | PUR? | | |
| L34 | 1 | S | ACARBOSE | (P) | AMMONIA | (P) | PURIFICATION | | |
| L35 | 1 | S | ACARBOSE | (P) | AMMONIA | (P) | PUR? | | |

L1 ANSWER 3 OF 3 MEDLINE on STN
ACCESSION NUMBER: 87190439 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3106037
TITLE: Purification and characterization of extracellular
alpha-amylase and glucoamylase from the yeast *Candida*
antarctica CBS 6678.
AUTHOR: De Mot R; Verachtert H
SOURCE: European journal of biochemistry / FEBS, (1987 May 4) Vol.
164, No. 3, pp. 643-54.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198706
ENTRY DATE: Entered STN: 3 Mar 1990
Last Updated on STN: 3 Mar 1990
Entered Medline: 25 Jun 1987

AB An alpha-amylase and a glucoamylase were purified to homogeneity from the culture fluid of beta-cyclodextrin-grown *Candida antarctica* CBS 6678 by protamine sulfate treatment, ammonium sulfate precipitation, gel filtration (Sephadex G-75 sf, Ultrogel ACA 54), DEAE-Sephacel chromatography, hydroxyapatite chromatography and affinity chromatography on acarbose--AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compositions. Their apparent relative molecular mass, sedimentation coefficient ($S_{20,w}$), isoelectric point, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2 and 57 degrees C, respectively, for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2 and 62 degrees C, respectively, for alpha-amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-molecular-mass substrates, including some raw starches. alpha-Amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both alpha-amylase (K_i less than 1 microM) and glucoamylase (K_i less than 0.1 microM), being more effective than Bay e 4609 (K_i less than 10 microM). Glucoamylase was selectivity and strongly inhibited by acarbose (K_i less than 0.1 microM). Activity of the latter enzyme was also affected by 1-deoxynojirimycin (K_i less than 1 mM), maltitol and amino alcohols (K_i less than 10 mM). Unlike alpha-amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being non-identical with the active site.

L1 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purification process for manufacturing a high pure acarbose relates to a process for preparing high pure acarbose from acarbose-containing fermentation broth. The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L1 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:209924 CAPLUS
DOCUMENT NUMBER: 106:209924
TITLE: Purification and characterization of extracellular
 α -amylase and glucoamylase from the yeast
Candida antarctica CBS 6678
AUTHOR(S): De Mot, Rene; Verachtert, Hubert
CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Heverlee,
B-3030, Belg.
SOURCE: European Journal of Biochemistry (1987), 164(3),
643-54
CODEN: EJBCAI; ISSN: 0014-2956
DOCUMENT TYPE: Journal
LANGUAGE: English

AB An α -amylase and a glucoamylase were purified to homogeneity from the culture fluid of β -cyclodextrin-grown *C. antarctica* CBS 6678 by protamine sulfate treatment, $(\text{NH}_4)_2\text{SO}_4$ precipitation, gel filtration (Sephadex G-75 sf, Ultrogel AcA 54), DEAE-Sephacel chromatog., hydroxyapatite chromatog., and affinity chromatog. on acarbose-AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compns. Their apparent relative mol. mass, sedimentation coefficient (s_{20,w°), pI, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7

S, 10.1,
1.74 $\text{cm}^2 \text{mg}^{-1}$, 4.2°, and 57°, resp., for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 $\text{cm}^2 \text{mg}^{-1}$, 4.2° and 62°, resp., for α -amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-mol.-mass substrates, including some raw starches. The α -amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both α -amylase ($K_i < 1 \mu\text{M}$) and glucoamylase ($K_i < 0.1 \mu\text{M}$), being more effective than Bay e 4609 ($K_i < 10 \mu\text{M}$). Glucoamylase was selectively and strongly inhibited by acarbose ($K_i < 0.1 \mu\text{M}$). Activity of the latter enzyme was also affected by 1-deoxynojirimycin ($K_i < 1 \text{mM}$), maltitol, and amino

alcs. ($K_i < 10$ mM). Unlike α -amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being nonidentical with the active site.

L1 ANSWER 3 OF 3 MEDLINE on STN
ACCESSION NUMBER: 87190439 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3106037
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antarctica CBS 6678.
AUTHOR: De Mot R; Verachtert H
SOURCE: European journal of biochemistry / FEBS, (1987 May 4) Vol.
164, No. 3, pp. 643-54.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198706
ENTRY DATE: Entered STN: 3 Mar 1990
Last Updated on STN: 3 Mar 1990
Entered Medline: 25 Jun 1987

AB An alpha-amylase and a glucoamylase were purified to homogeneity from the culture fluid of beta-cyclodextrin-grown *Candida antarctica* CBS 6678 by protamine sulfate treatment, ammonium sulfate precipitation, gel filtration (Sephadex G-75 sf, Ultrogel AcA 54), DEAE-Sephacel chromatography, hydroxyapatite chromatography and affinity chromatography on acarbose--AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compositions. Their apparent relative molecular mass, sedimentation coefficient ($S_{20,w}$), isoelectric point, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2 and 57 degrees C, respectively, for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2 and 62 degrees C, respectively, for alpha-amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-molecular-mass substrates, including some raw starches. alpha-Amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both alpha-amylase (K_i less than 1 microM) and glucoamylase (K_i less than 0.1 microM), being more effective than Bay e 4609 (K_i less than 10 microM). Glucoamylase was selectively and strongly inhibited by acarbose (K_i less than 0.1 microM). Activity of the latter enzyme was also affected by 1-deoxynojirimycin (K_i less than 1 mM), maltitol and amino alcohols (K_i less than 10 mM). Unlike alpha-amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being non-identical with the active site.

L5 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:924641 CAPLUS

DOCUMENT NUMBER: 142:155800

TITLE: Process for preparing α -glycosidase inhibitor derived from powdery silkworm

INVENTOR(S): Ahn, Mi Yeong; Kim, Eun Seon; Kim, Gwang Won; Kim, Ik Su; Kim, Jin Won; Lee, Hui Sam; Lee, Yong Gi; Ryu, Gang Seon; Seo, Su Won

PATENT ASSIGNEE(S): Republic of Korea Management : Rural Development Administration, S. Korea; Samsung Life Public Welfare Foundation Samsung Medical Center

SOURCE: Repub. Korean Kongkae Taeho Kongbo, No pp. given
CODEN: KRXXA7

DOCUMENT TYPE: Patent

LANGUAGE: Korean

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|-----------------|----------|
| ----- | ---- | ----- | ----- | ----- |
| KR 2002071334 | A | 20020912 | KR 2001-11449 | 20010306 |
| PRIORITY APPLN. INFO.: | | | KR 2001-11449 | 20010306 |

AB Provided is a process for preparing α -glycosidase inhibitor derived from powdery silkworm, which has the same decreasing effect of blood glucose level as acarbose which is a conventional therapeutic agent for diabetes. The process for preparing α -glycosidase inhibitor comprises the steps of: freeze-drying and powdering silkworm then adding 50% ethanol to extract it; performing cation exchange column chromatog. with the extract then followed by eluting it with 0.5N of NH₄OH and concentrating it; dissolving the concentrate in water then followed by performing anion exchange column chromatog. eluting with water and concentrating it; dissolving the concentrate in water then followed by cation exchange column chromatog., eluting with water and 0.5N of NH₄OH to obtain 5 fractions obtain one fraction resp.; and performing anion exchange column chromatog. with the second fraction among the 6 fractions to obtain pure 1-deoxynojirimycin fraction.

L6 ANSWER 1 OF 5 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:99343 CAPLUS
DOCUMENT NUMBER: 140:297257
TITLE: A novel α -glucosidase inhibitor from pine bark
AUTHOR(S): Kim, Yong-Mu; Wang, Myeong-Hyeon; Rhee, Hae-Ik
CORPORATE SOURCE: Division of Biotechnology, Kangwon National
University, Chuncheon, 200-701, S. Korea
SOURCE: Carbohydrate Research (2004), 339(3), 715-717
CODEN: CRBRAT; ISSN: 0008-6215
PUBLISHER: Elsevier
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Inhibitors of carbohydrate-hydrolyzing enzymes play an important role for the treatment of diabetes. To our knowledge, a number of inhibitors such as, 1-deoxynojirimycin, acarbose and voglibose have been identified as a result of screening of secondary metabolites up to now. In this note, we report the inhibitory effect on carbohydrate hydrolysis of ethanol exts. from more than 1400 species of plants with the aim of identifying a potential antihyperglycemic drug. *Pinus densiflora* bark exts. showed the highest inhibition activity against several carbohydrate-hydrolyzing enzymes.

REFERENCE COUNT: 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:58741 CAPLUS
DOCUMENT NUMBER: 140:246701
TITLE: Anti-diabetic effects of CCCA, CMES, and cordycepin from *Cordyceps militaris* and the immune responses in streptozotocin-induced diabetic mice
AUTHOR(S): Yun, Yunha; Han, Shinha; Lee, Seungjeong; Ko, Sung Kwon; Lee, Chong-kil; Ha, Nam-joo; Kim, Kyungjae
CORPORATE SOURCE: Department of Pharmacy, Sahmyook University, Seoul, 139-742, S. Korea
SOURCE: Natural Product Sciences (2003), 9(4), 291-298
CODEN: NPSCFB; ISSN: 1226-3907
PUBLISHER: Korean Society of Pharmacognosy
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Anti-diabetic effect of various fractions of *Cordyceps militaris* (CM), CCCA (crude cordycepin containing adenosine), CMES (ethanol soluble supernatant), and cordycepin were evaluated in streptozotocin (STZ) induced diabetic mice. CMES showed potent inhibitory activity of 34.7% in starch-loaded mice (2 g/kg) while acarbose as a pos. standard exhibited 37.8% of inhibition rate. After 3 days administration (50 mg/kg), CMES reduced blood glucose level by 35.5% (acarbose, 37.2%). However CCCA, cordycepin, and tryptophan showed no significance. After 7 days administrations for the long-term usage of these drugs, CMES (50 mg/kg), cordycepin (0.2 mg/kg), and acarbose (10 mg/kg) dramatically reduced blood glucose level (inhibition ratio: 46.9%, 48.4% and 37.5% resp.). CCCA that has high contents of cordycepin (0.656 mg/4 mg) did not have influence on reducing blood glucose level. The proliferation of splenocytes and peritoneal macrophages derived from STZ-induced diabetic mice administered samples were evaluated out by addition of mitogens to see the stability of the usage of these herbal medicines. Proliferation of T-lymphocyte was significantly decreased; while NO production was increased more than two fold to STZ control in the cordycepin-administered group. Proliferation of macrophages and NO production were significantly decreased in CMES administered group. Changes of serum enzyme levels of glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT) were also evaluated. Cordycepin-administered group appeared to have higher levels than control in both enzymes, but it was not significant as compared to

acarbose. We conclude that CMES and cordycepin may be useful tools in the control of blood glucose level in diabetes and promising new drug as an anti-hyperglycemic agent without defects of immune responses and other side effects.

REFERENCE COUNT: 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1999:37149 CAPLUS

DOCUMENT NUMBER: 130:233533

TITLE: Acarbose alone or in combination with ethanol potentiates the hepatotoxicity of carbon tetrachloride and acetaminophen in rats

AUTHOR(S): Wang, Pei-Yu; Kaneko, Takashi; Wang, Yuan; Sato, Akio

CORPORATE SOURCE: Department of Environmental Health, Medical University of Yamanashi, Yamanashi, 409-3898, Japan

SOURCE: Hepatology (Philadelphia) (1999), 29(1), 161-165

CODEN: HPTLD9; ISSN: 0270-9139

PUBLISHER: W. B. Saunders Co.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Acarbose reduces the absorption of monosaccharides derived from dietary carbohydrates, which play an important role in the metabolism and toxicity of some chemical compds. We studied the effects of acarbose on the hepatotoxicity of carbon tetrachloride (CCl₄) and acetaminophen (AP) in rats, both of which exert their toxic effects through bioactivation associated with cytochrome P 450 2E1 (CYP2E1). Male Sprague-Dawley rats were kept on a daily ration (20 g) of powdered chow diet containing 0, 20, 40, or 80 mg/100 g of acarbose, with drinking water containing 0% or 10% of ethanol (volume/volume). Three weeks later, the rats were either killed for an in vitro metabolism study or challenged with 0.50 g/kg CCl₄ orally or 0.75 g/kg AP i.p. The ethanol increased the hepatic microsomal CYP2E1 level and the rate of dimethylnitrosamine (DMN) demethylation. The 40- or 80-mg/100 g acarbose diet, which alone increased the CYP2E1 level and the rate of DMN demethylation, augmented the enzyme induction by ethanol. The 40- or 80-mg/100 g acarbose diet alone potentiated CCl₄ and AP hepatotoxicity, as evidenced by significantly increased levels of both alanine transaminase (ALT) and aspartate transaminase (AST) in the plasma of rats pretreated with acarbose. Ethanol alone also potentiated the toxicity of both chems. When the 40- or 80-mg/100 g acarbose diet was combined with ethanol, the ethanol-induced potentiation of CCl₄ and AP hepatotoxicity was augmented. Our study demonstrated that high doses of acarbose, alone or in combination with ethanol, can potentiate CCl₄ and AP hepatotoxicity in rats by inducing hepatic CYP2E1.

REFERENCE COUNT: 52 THERE ARE 52 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L6 ANSWER 4 OF 5 MEDLINE on STN

ACCESSION NUMBER: 2004122257 MEDLINE

DOCUMENT NUMBER: PubMed ID: 15013410

TITLE: A novel alpha-glucosidase inhibitor from pine bark.

AUTHOR: Kim Yong-Mu; Wang Myeong-Hyeon; Rhee Hae-Ik

CORPORATE SOURCE: Division of Biotechnology, Kangwon National University, Chunchon 200-701, South Korea.

SOURCE: Carbohydrate research, (2004 Feb 25) Vol. 339, No. 3, pp. 715-7.

Journal code: 0043535. ISSN: 0008-6215.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200411

ENTRY DATE: Entered STN: 12 Mar 2004
Last Updated on STN: 5 Nov 2004
Entered Medline: 4 Nov 2004

AB Inhibitors of carbohydrate-hydrolysing enzymes play an important role for the treatment of diabetes. To our knowledge, a number of inhibitors such as, 1-deoxynojirimycin, acarbose and voglibose have been identified as a result of screening of secondary metabolites up to now. In this note, we report the inhibitory effect on carbohydrate hydrolysis of ethanol extracts from more than 1400 species of plants with the aim of identifying a potential antihyperglycemic drug. *Pinus densiflora* bark extracts showed the highest inhibition activity against several carbohydrate-hydrolysing enzymes.

L6 ANSWER 5 OF 5 MEDLINE on STN
ACCESSION NUMBER: 1999081692 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9862862
TITLE: Acarbose alone or in combination with ethanol potentiates the hepatotoxicity of carbon tetrachloride and acetaminophen in rats.
AUTHOR: Wang P Y; Kaneko T; Wang Y; Sato A
CORPORATE SOURCE: Department of Environmental Health, Medical University of Yamanashi, Tamaho, Yamanashi, Japan.
SOURCE: Hepatology (Baltimore, Md.), (1999 Jan) Vol. 29, No. 1, pp. 161-5.
Journal code: 8302946. ISSN: 0270-9139.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199902
ENTRY DATE: Entered STN: 16 Feb 1999
Last Updated on STN: 17 Dec 2002
Entered Medline: 3 Feb 1999

AB Acarbose reduces the absorption of monosaccharides derived from dietary carbohydrates, which play an important role in the metabolism and toxicity of some chemical compounds. We studied the effects of acarbose on the hepatotoxicity of carbon tetrachloride (CCl₄) and acetaminophen (AP) in rats, both of which exert their toxic effects through bioactivation associated with cytochrome P450 2E1 (CYP2E1). Male Sprague-Dawley rats were kept on a daily ration (20 g) of powdered chow diet containing 0, 20, 40, or 80 mg/100 g of acarbose, with drinking water containing 0% or 10% of ethanol (vol/vol). Three weeks later, the rats were either killed for an in vitro metabolism study or challenged with 0.50 g/kg CCl₄ orally or 0.75 g/kg AP intraperitoneally. The ethanol increased the hepatic microsomal CYP2E1 level and the rate of dimethylnitrosamine (DMN) demethylation. The 40- or 80-mg/100 g acarbose diet, which alone increased the CYP2E1 level and the rate of DMN demethylation, augmented the enzyme induction by ethanol. The 40- or 80-mg/100 g acarbose diet alone potentiated CCl₄ and AP hepatotoxicity, as evidenced by significantly increased levels of both alanine transaminase (ALT) and aspartate transaminase (AST) in the plasma of rats pretreated with acarbose. Ethanol alone also potentiated the toxicity of both chemicals. When the 40- or 80-mg/100 g acarbose diet was combined with ethanol, the ethanol-induced potentiation of CCl₄ and AP hepatotoxicity was augmented. Our study demonstrated that high doses of acarbose, alone or in combination with ethanol, can potentiate CCl₄ and AP hepatotoxicity in rats by inducing hepatic CYP2E1.

L7 ANSWER 3 OF 3 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose (BAY g-5421)
affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L7 ANSWER 3 OF 3 MEDLINE on STN
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AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L7 ANSWER 3 OF 3 MEDLINE on STN
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AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L7 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purifn. process for manufacturing a high pure acarbose
relates to a process for preparing high pure acarbose from
acarbose-containing fermentation broth. The acarbose was purified
through steps of alc. precipitation, a strongly acidic cation exchanger
chromatog.
and an immobilized enzyme affinity chromatog.
Acarbose is generally applied in treating diabetes.

L7 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS
DOCUMENT NUMBER: 105:167504
TITLE: Purification of glucoamylase by acarbose (BAY g-5421)
affinity chromatography
AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.
CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA
SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3),
201-9
CODEN: BABIEC; ISSN: 0885-4513
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was purified
from com. available, impure enzyme prepns. by affinity
chromatog. on acarbose (II) columns. Up to 2 mg I was bound
without leakage to a 1-mL affinity gel column possessing a
covalently linked II ligand (1 mg II/g wet gel), and the bound
enzyme was specifically released by irrigation of the column with
a solution of maltose. A complete cycle of purifn. was
accomplished in .apprx.8 h. Both I activities were recovered in >80%
yield, free of α -amylase activity and possessing specific activities
comparable to those of prepns. obtained by time-consuming, multistep
procedures involving several ion-exchange and hydrophobic column
fractionations. Thus, II affinity chromatog. provides a general
method for the rapid and efficient purifn. of I, and appears to
be ideally suited for scale-up for the com. purifn. of these
enzymes.

L7 ANSWER 3 OF 3 MEDLINE on STN

ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose (BAY g-5421)
affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)

SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol. 8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L8 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

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Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

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| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

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acarbose-containing fermn. broth. The
acarbose was purified through steps of alc. precipitation, a strongly
acidic cation exchanger chromatog. and an immobilized enzyme affinity
chromatog. Acarbose is generally applied in treating diabetes.

L8 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:928233 CAPLUS
DOCUMENT NUMBER: 138:3755
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Hung.
SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S.
Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|--|----------|-----------------|-------------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | US 2001-924271 | A2 20010807 |

AB The present invention relates to a novel process for the preparation of
acarbose. Said process comprises the steps of: acidifying a fermentation broth
containing an acarbose; removing particulates from the fermentation broth;
adsorbing
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L8 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

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PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

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L8 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:928233 CAPLUS
DOCUMENT NUMBER: 138:3755
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SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S.
Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|--|----------|-----------------|-------------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | US 2001-924271 | A2 20010807 |

AB The present invention relates to a novel process for the preparation of
acarbose. Said process comprises the steps of: acidifying a fermentation broth
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adsorbing
the acarbose on a cation-exchanger in the presence of an anion of a weak

acid; eluting the acarbose from the cation-exchanger with at least one of a sodium chloride solution and a salt solution; precipitating the acarbose with a solvent; and recovering the precipitated acarbose.

L8 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 2002:123021 CAPLUS
DOCUMENT NUMBER: 136:182542
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.
SOURCE: PCT Int. Appl., 24 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|------------|
| WO 2002012256 | A1 | 20020214 | WO 2001-US24729 | 20010807 |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
| AU 2001084741 | A5 | 20020218 | AU 2001-84741 | 20010807 |
| EP 1309601 | A1 | 20030514 | EP 2001-963821 | 20010807 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | WO 2001-US24729 | W 20010807 |

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REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

acid; eluting the acarbose from the cation-exchanger with at least one of a sodium chloride solution and a salt solution; precipitating the acarbose with a solvent; and recovering the precipitated acarbose.

L8 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 2002:123021 CAPLUS
DOCUMENT NUMBER: 136:182542
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.
SOURCE: PCT Int. Appl., 24 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
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| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
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| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | WO 2001-US24729 | W 20010807 |

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REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 2 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN
 ACCESSION NUMBER: 2002:928233 CAPLUS
 DOCUMENT NUMBER: 138:3755
 TITLE: Method for purification of acarbose
 INVENTOR(S): Keri, Vilmos; Deak, Lajos
 PATENT ASSIGNEE(S): Hung.
 SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S.
 Ser. No. 924,271.
 CODEN: USXXCO
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 2
 PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|------|----------|-----------------|----------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
 CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH,
 GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
 LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH,
 PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ,
 UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU,
 TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH,
 CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR,
 BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-223492P P 20000807
 US 2001-924271 A2 20010807

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| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

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L9 ANSWER 2 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

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DOCUMENT NUMBER: 138:3755
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PATENT ASSIGNEE(S): Hung.
SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S.
Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

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| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | US 2001-924271 | A2 20010807 |

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L9 ANSWER 3 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:854107 CAPLUS

DOCUMENT NUMBER: 138:149416

TITLE: Isolation, characterization and inhibition by acarbose of the α -amylase from *Lactobacillus fermentum*: comparison with *Lb. manihotivorans* and *Lb. plantarum* amylases

AUTHOR(S): Talamond, P.; Desseaux, V.; Moreau, Y.; Santimone, M.; Marchis-Mouren, G.

CORPORATE SOURCE: IMRN, Institut Meditteraneen de Recherche en Nutrition, Faculte des Sciences et Techniques de St Jerome, Universite d'Aix-Marseille, Marseille, F-13397, Fr.

SOURCE: Comparative Biochemistry and Physiology, Part B: Biochemistry & Molecular Biology (2002), 133B(3), 351-360

CODEN: CBPBB8; ISSN: 1096-4959

PUBLISHER: Elsevier Science Inc.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Extracellular α -amylase from *Lactobacillus fermentum* (FERMENTA) was purified by glycogen precipitation and ion exchange chromatog. The purifn. was approx. 28-fold with a 27% yield. The FERMENTA mol. mass (106 000 Da) is in the same range as the ones determined for *L. amylovorus* (AMYLOA), *L. plantarum* (PLANTAA) and *L. manihotivorans* (MANIHOA) α -amylases. The amino acid composition of FERMENTA differs from the other lactobacilli considered here, but however, indicates that the peptidic sequence contains two equal parts: the N-terminal catalytic part; and the C-terminal repeats. The isoelec. point of FERMENTA, PLANTAA, MANIHOA are approx. the same (3.6). The FERMENTA optimum pH (5.0) is slightly more acidic and the optimum temperature is lower (40°). Raw starch hydrolysis catalyzed by all three amylases liberates maltotriose and maltotetraose. Maltose is also produced by FERMENTA and MANIHOA. Maltohexaose FERMENTA catalyzed hydrolysis produces maltose and maltotriose. Finally, kinetic studies of FERMENTA, PLANTAA and MANIHOA using amylose as a substrate and acarbose as an inhibitor, were carried out. Statistical anal. of kinetic data, expressed using a general velocity equation and assuming rapid equilibrium, showed that: (1) in the absence of inhibitor k_{cat}/K_m are, resp., $1 + 109$, $12.6 + 109$ and $3.2 + 109 \text{ s}^{-1} \text{ M}^{-1}$; and (2) the inhibition of FERMENTA is of the mixed non-competitive type ($K_{li}=5.27 \mu\text{M}$; $L_{li}=1.73 \mu\text{M}$) while the inhibition of PLANTAA and MANIHOA is of the uncompetitive type ($L_{li}=1.93 \mu\text{M}$ and $1.52 \mu\text{M}$, resp.). Whatever the inhibition type, acarbose is a strong inhibitor of these *Lactobacillus* amylases. These results indicate that, as found in porcine and barley amylases, *Lactobacillus* amylases contain in addition to the active site, a soluble carbohydrate (substrate or product) binding site.

REFERENCE COUNT: 25 THERE ARE 25 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 4 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:123021 CAPLUS

DOCUMENT NUMBER: 136:182542

TITLE: Method for purification of acarbose

INVENTOR(S): Keri, Vilmos; Deak, Lajos

PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.

SOURCE: PCT Int. Appl., 24 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|--|----------|-----------------|------------|
| WO 2002012256 | A1 | 20020214 | WO 2001-US24729 | 20010807 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| AU 2001084741 | A5 | 20020218 | AU 2001-84741 | 20010807 |
| EP 1309601 | A1 | 20030514 | EP 2001-963821 | 20010807 |
| R: | AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | WO 2001-US24729 | W 20010807 |

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of hydrochloric acid and the weak acid; precipitating the acarbose with a solvent; and separating the acarbose crystals.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L9 ANSWER 5 OF 14 CAPLUS COPYRIGHT 2006 ACS on STM
ACCESSION NUMBER: 1999:126912 CAPLUS
DOCUMENT NUMBER: 130:139581
TITLE: A process for the purification of acarbose
INVENTOR(S): Lawton, Carl William; Swartz, Randall Wolfe
PATENT ASSIGNEE(S): University of Massachusetts Lowell, USA
SOURCE: PCT Int. Appl., 11 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|--|----------|-----------------|----------|
| WO 9907720 | A2 | 19990218 | WO 1998-IB1188 | 19980803 |
| WO 9907720 | A3 | 19990415 | | |
| W: | AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG | | | |
| AU 9883542 | A1 | 19990301 | AU 1998-83542 | 19980803 |
| EP 1003761 | A2 | 20000531 | EP 1998-933857 | 19980803 |
| R: | AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI | | | |
| JP 2001512738 | T2 | 20010828 | JP 2000-506222 | 19980803 |

PRIORITY APPLN. INFO.:

IT 1997-MI1880

A 19970805

WO 1998-IB1188

W 19980803

AB The present invention discloses a process for the purifn. of acarbose comprising loading a pre-purified acarbose solution on a chromatog. column packed with a non-aromatic strong acid cation exchanger which is hydrophilic and has high mass transfer and subsequent elution.

L9 ANSWER 6 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1993:423633 CAPLUS

DOCUMENT NUMBER: 119:23633

TITLE: Production, purification and characterization of the catalytic domain of glucoamylase from *Aspergillus niger*

AUTHOR(S): Stoffer, Bjarne; Frandsen, Torben P.; Busk, Peter K.; Schneider, Palle; Svendsen, Ib; Svensson, Birte

CORPORATE SOURCE: Dep. Chem., Carlsberg Lab., Valby, DK-2500, Den.

SOURCE: Biochemical Journal (1993), 292(1), 197-202

CODEN: BIJOAK; ISSN: 0306-3275

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The catalytic domain of glucoamylases G1 and G2 from *A. niger* was produced in vitro in high yield by limited proteolysis using either subtilisin Novo or subtilisin Carlsberg. Purifn. by affinity chromatog. on an acarbose-Sepharose column followed by ion-exchange chromatog. on HiLoad Q-Sepharose led to separation of a number of structurally closely related forms of domain. The cleavage occurred primarily between Val-470 and Ala-471 as indicated by C-terminal sequencing, whereas the N-terminus was intact. Subtilisin Carlsberg, in addition, produces a type of domain which was hydrolyzed before Ser-444, an O-glycosylated residue. This left the fragment, Ser-444-Val-470, disulfide-bonded to the large N-terminal part of the catalytic domain. Subtilisin Novo, in contrast, tended to yield a minor fraction of forms extending approx. 30-40 amino-acid residues beyond Val-470. The thermostability was essentially the same for the single-chain catalytic domain and the original glucoamylases G1 and G2, whereas the catalytic domain cleaved between Ser-443 and Ser-444 was less thermostable. For both types of domain, the kinetic parameters, K_m and k_{cat} , for the hydrolysis of maltose were very close to the values found for glucoamylases G1 and G2.

L9 ANSWER 7 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1989:403061 CAPLUS

DOCUMENT NUMBER: 111:3061

TITLE: Single step affinity chromatographic purification of human α -amylase from aspirated duodenal juice and its application in the measurement of pancreatic α -amylase synthesis rates in man

AUTHOR(S): Ogden, J. M.; O'Keefe, S. J. D.; Ehlers, M. R. W.; Kirsch, R. E.; Marks, I. N.

CORPORATE SOURCE: Gastrointest. Clin., Groote Schuur Hosp., Observatory, 7925, S. Afr.

SOURCE: Clinica Chimica Acta (1989), 180(2), 129-39

CODEN: CCATAR; ISSN: 0009-8981

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Human α -amylase was purified from aspirated duodenal juice to electrophoretic homogeneity in a single step by affinity chromatog. with the competitive inhibitor, acarbose ($IC_{50} = 1.22 \mu M$), as ligand. Duodenal juice was applied to an agarose resin to which acarbose had been coupled covalently via a 1.9-nm spacer group. Pure α -amylase, eluted with free acarbose, had a mol. weight of 55,000, and isoelectrofocusing revealed the presence of 6 isoenzymes with pI values of 7.3, 6.8, 6.7, 6.5, 6.4, and 6.3 all of which possessed

amylase activity based on pos. starch/iodine staining. The potential usefulness of this 1-step purifn. procedure in the measurement of pancreatic α -amylase synthesis rates was evaluated in 2 control patients with nonpancreatic disease. Aspirated duodenal juice was obtained during a pulse/continuous i.v. 4-h infusion of [14C]leucine together with secretin and pancreozymin, and α -amylase was purified using the protocol. Pancreatic α -amylase synthesis rates were determined from the rate of incorporation of [14C]leucine into α -amylase; values of 4.4 and 5.1 h were obtained for the 2 control patients.

L9 ANSWER 8 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:524834 CAPLUS

DOCUMENT NUMBER: 109:124834

TITLE: Effective purification of glucoamylase in koji, a solid culture of *Aspergillus oryzae* on steamed rice, by affinity chromatography using an immobilized acarbose (BAY g-5421)

AUTHOR(S): Ono, Kazuhisa; Shigeta, Seiko; Oka, Satoru

CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan

SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1707-14

CODEN: ABCHA6; ISSN: 0002-1369

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Glucoamylase (GA) was purified from koji, a solid culture of *A. oryzae* on steamed rice, by extraction with 1% NaCl solution, precipitation with EtOH, and acarbose

affinity chromatog. The purified enzyme was homogeneous on gel filtration, PAGE and SDS-PAGE, ultracentrifugation, and IEF. The enzyme released β -glucose as a sole product from soluble starch and maltooligosaccharides. The other common and inherent features of GAs were also confirmed in the GA from *A. oryzae*. The enzyme was a glycoprotein containing .apprx.4.8% glucosamine and 7.8% neutral saccharides.

L9 ANSWER 9 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:497062 CAPLUS

DOCUMENT NUMBER: 107:97062

TITLE: Purification of acarbose via chromatography on weakly acidic cation exchange resins

INVENTOR(S): Rauenbusch, Erich

PATENT ASSIGNEE(S): Bayer A.-G., Fed. Rep. Ger.

SOURCE: Ger. Offen., 7 pp.

CODEN: GWXXBX

DOCUMENT TYPE: Patent

LANGUAGE: German

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|----------|
| DE 3543999 | A1 | 19870619 | DE 1985-3543999 | 19851213 |
| EP 226121 | A2 | 19870624 | EP 1986-116773 | 19861202 |
| EP 226121 | A3 | 19890322 | | |
| EP 226121 | B1 | 19920122 | | |
| R: AT, BE, CH, DE, ES, FR, GB, IT, LI, NL, SE | | | | |
| AT 71951 | E | 19920215 | AT 1986-116773 | 19861202 |
| ES 2038591 | T3 | 19930801 | ES 1986-116773 | 19861202 |
| JP 62155288 | A2 | 19870710 | JP 1986-292667 | 19861210 |
| JP 2502551 | B2 | 19960529 | | |
| US 4904769 | A | 19900227 | US 1986-940713 | 19861211 |
| CA 1288768 | A1 | 19910910 | CA 1986-525014 | 19861211 |
| DK 8605986 | A | 19870614 | DK 1986-5986 | 19861212 |

| | | | | |
|-------------|----|----------|----------------|----------|
| DK 164870 | B | 19920831 | | |
| DK 164870 | C | 19930111 | | |
| HU 43083 | A2 | 19870928 | HU 1986-5203 | 19861212 |
| HU 196219 | B | 19881028 | | |
| CN 86108259 | A | 19870729 | CN 1986-108259 | 19861213 |
| CN 1013866 | B | 19910911 | | |
| JP 08245683 | A2 | 19960924 | JP 1996-34402 | 19960129 |
| JP 2628853 | B2 | 19970709 | | |

PRIORITY APPLN. INFO.:

| | | |
|-----------------|---|----------|
| DE 1985-3543999 | A | 19851213 |
| EP 1986-116773 | A | 19861202 |

AB Acarbose (I) containing <10% impurity (excluding H₂O) is prepared by passing a 1-20% solution of I at pH 4-7 through a column packed with a dextran-, agarose-, or cellulose-based carboxylic acid ion exchanger optionally containing a polyamide. A solution of 9.2 g impure I in .apprx.40 mL H₂O adjusted to pH 4.7 was added to a 2.6 + 34 cm column containing CM-Sephadex C 25 previously treated with a NaOAc buffer and flushed with H₂O. I was eluted with H₂O at 100 mL/h to give 5.87 g of 93% pure I.

L9 ANSWER 10 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS

DOCUMENT NUMBER: 105:167504

TITLE: Purification of glucoamylase by acarbose (BAY g-5421) affinity chromatography

AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.

CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA

SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3), 201-9
CODEN: BABIEC; ISSN: 0885-4513

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was purified from com. available, impure enzyme preps. by affinity chromatog. on acarbose (II) columns. Up to 2 mg I was bound without leakage to a 1-mL affinity gel column possessing a covalently linked II ligand (1 mg II/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purifn. was accomplished in .apprx.8 h. Both I activities were recovered in >80% yield, free of α -amylase activity and possessing specific activities comparable to those of preps. obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, II affinity chromatog. provides a general method for the rapid and efficient purifn. of I, and appears to be ideally suited for scale-up for the com. purifn. of these enzymes.

L9 ANSWER 11 OF 14 MEDLINE on STN

ACCESSION NUMBER: 2002739743 MEDLINE

DOCUMENT NUMBER: PubMed ID: 12431403

TITLE: Isolation, characterization and inhibition by acarbose of the alpha-amylase from *Lactobacillus fermentum*: comparison with *Lb. manihotivorans* and *Lb. plantarum* amylases.

AUTHOR: Talamond P; Desseaux V; Moreau Y; Santimone M; Marchis-Mouren G

CORPORATE SOURCE: Faculte des Sciences et Techniques de St Jerome, Universite d'Aix-Marseille, IMRN, Institut Mediterranee de Recherche en Nutrition, av Escadrille Normandie-Niemen, F-13397, Marseille cedex 20, France.

SOURCE: Comparative biochemistry and physiology. Part B, Biochemistry & molecular biology, (2002 Nov) Vol. 133, No. 3, pp. 351-60.

Journal code: 9516061. ISSN: 1096-4959.

PUB. COUNTRY: England: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200307
ENTRY DATE: Entered STN: 31 Dec 2002
Last Updated on STN: 29 Jul 2003
Entered Medline: 28 Jul 2003

AB Extracellular alpha-amylase from *Lactobacillus fermentum* (FERMENTA) was purified by glycogen precipitation and ion exchange chromatography. The purification was approximately 28-fold with a 27% yield. The FERMENTA molecular mass (106,000 Da) is in the same range as the ones determined for *L. amylovorus* (AMYLOA), *L. plantarum* (PLANTAA) and *L. manihotivorus* (MANIHOA) alpha-amylases. The amino acid composition of FERMENTA differs from the other lactobacilli considered here, but however, indicates that the peptidic sequence contains two equal parts: the N-terminal catalytic part; and the C-terminal repeats. The isoelectric point of FERMENTA, PLANTAA, MANIHOA are approximately the same (3.6). The FERMENTA optimum pH (5.0) is slightly more acidic and the optimum temperature is lower (40 degrees C). Raw starch hydrolysis catalyzed by all three amylases liberates maltotriose and maltotetraose. Maltose is also produced by FERMENTA and MANIHOA. Maltohexaose FERMENTA catalyzed hydrolysis produces maltose and maltotriose. Finally, kinetics of FERMENTA, PLANTAA and MANIHOA using amylose as a substrate and acarbose as an inhibitor, were carried out. Statistical analysis of kinetic data, expressed using a general velocity equation and assuming rapid equilibrium, showed that: (1) in the absence of inhibitor $k(\text{cat})/K_m$ are, respectively, 1×10^9 , 12.6×10^9 and $3.2 \times 10^9 \text{ s}^{-1} \text{ M}^{-1}$; and (2) the inhibition of FERMENTA is of the mixed non-competitive type ($K(1i)=5.27 \text{ microM}$; $L(1i)=1.73 \text{ microM}$) while the inhibition of PLANTAA and MANIHOA is of the uncompetitive type ($L(1i)=1.93 \text{ microM}$ and 1.52 microM , respectively). Whatever the inhibition type, acarbose is a strong inhibitor of these *Lactobacillus* amylases. These results indicate that, as found in porcine and barley amylases, *Lactobacillus* amylases contain in addition to the active site, a soluble carbohydrate (substrate or product) binding site.

L9 ANSWER 12 OF 14 MEDLINE on STN
ACCESSION NUMBER: 93277459 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8503847
TITLE: Production, purification and characterization of the catalytic domain of glucoamylase from *Aspergillus niger*.
AUTHOR: Stoffer B; Frandsen T P; Busk P K; Schneider P; Svendsen I; Svensson B
CORPORATE SOURCE: Carlsberg Laboratory, Department of Chemistry, Valby, Copenhagen, Denmark.
SOURCE: The Biochemical journal, (1993 May 15) Vol. 292 (Pt 1), pp. 197-202.
Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199306
ENTRY DATE: Entered STN: 16 Jul 1993
Last Updated on STN: 16 Jul 1993
Entered Medline: 25 Jun 1993

AB The catalytic domain of glucoamylases G1 and G2 from *Aspergillus niger* is produced in vitro in high yield by limited proteolysis using either subtilisin Novo or subtilisin Carlsberg. Purification by affinity chromatography on an acarbose-Sepharose column followed by ion-exchange chromatography on HiLoad Q-Sepharose leads to separation of a number of structurally closely related forms of domain. The cleavage occurs primarily between Val-470 and Ala-471 as indicated by C-terminal sequencing, whereas the N-terminus is intact. Subtilisin Carlsberg, in addition, produces a type of domain

which is hydrolysed before Ser-444, an O-glycosylated residue. This leaves the fragment Ser-444-Val-470 disulphide-bonded to the large N-terminal part of the catalytic domain. Subtilisin Novo, in contrast, tends to yield a minor fraction of forms extending approx. 30-40 amino-acid residues beyond Val-470. The thermostability is essentially the same for the single-chain catalytic domain and the original glucoamylases G1 and G2, whereas the catalytic domain cut between Ser-443 and Ser-444 is less thermostable. For both types of domain the kinetic parameters, K_m and k_{cat} , for hydrolysis of maltose are very close to the values found for glucoamylases G1 and G2.

L9 ANSWER 13 OF 14 MEDLINE on STN
ACCESSION NUMBER: 89275526 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2786460
TITLE: Single step affinity chromatographic purification of human alpha-amylase from aspirated duodenal juice and its application in the measurement of pancreatic alpha-amylase synthesis rates in man.
AUTHOR: Ogden J M; O'Keefe S J; Ehlers M R; Kirsch R E; Marks I N
CORPORATE SOURCE: Gastrointestinal Clinic, Groote Schuur Hospital, University of Cape Town Medical School, Republic of South Africa.
SOURCE: Clinica chimica acta; international journal of clinical chemistry, (1989 Feb 28) Vol. 180, No. 2, pp. 129-39. Journal code: 1302422. ISSN: 0009-8981.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198907
ENTRY DATE: Entered STN: 9 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 20 Jul 1989

AB Human alpha-amylase was purified from aspirated duodenal juice to electrophoretic homogeneity in a single step by affinity chromatography with the competitive inhibitor acarbose ($IC_{50} = 1.22 \text{ } \mu\text{mol/l}$) as ligand. Duodenal juice was applied to an agarose resin to which acarbose had been coupled covalently via a 1.9 nm spacer group. Pure alpha-amylase, eluted with free acarbose, had a molecular mass of 55,000, and isoelectrofocusing revealed the presence of six isozymes with pI values of 7.3, 6.8, 6.7, 6.5, 6.4 and 6.3, all of which possessed amylase activity based on positive starch/iodine staining. The potential usefulness of this one-step purification procedure in the measurement of pancreatic alpha-amylase synthesis rates was evaluated in two control patients with non-pancreatic disease. Aspirated duodenal juice was obtained during a pulse/continuous intravenous 4 h infusion of [^{14}C]leucine together with secretin and pancreozymin, and alpha-amylase purified using our protocol. Pancreatic alpha-amylase synthesis rates were determined from the rate of incorporation of [^{14}C]leucine into alpha-amylase; values of 4.4 and 5.1 h were obtained for the two control patients.

L9 ANSWER 14 OF 14 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose (BAY g-5421) affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol. 8, No. 2-3, pp. 201-9. Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L11 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS

DOCUMENT NUMBER: 143:6386

TITLE: Purification process for manufacturing a high purity acarbose

INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu, Chi-Sheng

PATENT ASSIGNEE(S): Taiwan

SOURCE: U.S. Pat. Appl. Publ., 10 pp.

CODEN: USXXCO

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| ----- | --- | ----- | ----- | ----- |
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purification process for manufacturing a high pure acarbose relates to a process for preparing high pure acarbose from acarbose -containing fermentation broth. The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L14 ANSWER 1 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:1112573 CAPLUS

DOCUMENT NUMBER: 144:384721

TITLE: Enzymatic characterization of a maltogenic amylase from *Lactobacillus gasseri* ATCC 33323 expressed in *Escherichia coli*

AUTHOR(S): Oh, Ko-Woon; Kim, Myo-Jeong; Kim, Hae-Yeong; Kim, Byung-Yong; Baik, Moo-Yeol; Auh, Joong-Hyuck; Park, Cheon-Seok

CORPORATE SOURCE: Department of Food Science and Biotechnology, Institute of Life Sciences and Resources, KyungHee University, Yongin, 449-701, S. Korea

SOURCE: FEMS Microbiology Letters (2005), 252(1), 175-181
CODEN: FMLED7; ISSN: 0378-1097

PUBLISHER: Elsevier B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A gene corresponding to a maltogenic amylase (EC 3.2.1.133) (I) in *L. gasseri* ATCC 33323 was cloned and expressed in *Escherichia coli*. Recombinant I was efficiently purified 24.3-fold by 1-step Ni-NTA affinity chromatog. The final yield and specific activity of purified recombinant I was 68% and 58.7 U/mg, resp. Purified I exhibited optimal activity for β -cyclodextrin (β -CD) hydrolysis at 55° and pH 5. The relative hydrolytic activities of I with β -CD, soluble starch, and pullulan was 8:1:1.9. I was strongly inhibited by most metal cations, especially Zn²⁺, Fe²⁺, and Co²⁺, and by EDTA. *L. gasseri* I possessed some unusual properties distinguishable from typical I enzymes, such as being in a tetrameric form, exhibiting hydrolyzing activity toward the α -(1,6)-glycosidic linkage, and being inhibited by acarbose.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 2 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2003:589018 CAPLUS

DOCUMENT NUMBER: 140:108581

TITLE: Inhibitory effects of human and porcine α -amylase on CCK-8-stimulated lipase secretion of Isolated Rat Pancreatic Acini

AUTHOR(S): Jonas, Ludwig; Mikkat, Ulrike; Lehmann, Renate; Schareck, Wolfgang; Walzel, Hermann; Schroeder, Werner; Lopp, Hilja; Puessa, Toenu; Toomik, Peeter

CORPORATE SOURCE: Faculty of Medicine, Department of Pathology, University of Rostock, Germany

SOURCE: Pancreatology (2003), 3(4), 342-348
CODEN: PANCC2; ISSN: 1424-3903

PUBLISHER: S. Karger AG

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Previously we have demonstrated inhibitory effects of the plant lectin wheat germ agglutinin (WGA) on 125I-CCK-8 binding to pancreatic AR42J cells as well as on CCK-8-stimulated Ca²⁺ release and α -amylase secretion of rat pancreatic acini or acinar cells. Therefore, it is entirely conceivable that α -amylase having several lectin-like carbohydrate recognition domains can modulate the CCK-8 stimulated lipase secretion. Human α -amylase, purified from pancreatic juice by affinity chromatog. to homogeneity, and com. porcine pancreatic α -amylase inhibit CCK-8-stimulated lipase secretion of rat pancreatic acini in a concentration-dependent manner. Acarbose, a specific inhibitor of α -amylase, was without effect on CCK-8-induced cellular lipase secretion. The data presented here provide evidence for a regulatory function of α -amylase in CCK-8-stimulated pancreatic secretion.

REFERENCE COUNT: 22 THERE ARE 22 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 3 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1998:241502 CAPLUS

DOCUMENT NUMBER: 129:51181

TITLE: Alpha-glucosidase from the hepatopancreas of the shrimp, *Penaeus vannamei* (Crustacea-Decapoda)

AUTHOR(S): Le Chevalier, Patrick; Van Wormhoudt, Alain

CORPORATE SOURCE: Institut Universitaire de Technologie, Quimper, 29334, Fr.

SOURCE: Journal of Experimental Zoology (1998), 280(6), 384-394

CODEN: JEZOAO; ISSN: 0022-104X

PUBLISHER: Wiley-Liss, Inc.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB *Penaeus vannamei* is an omnivorous species, and it can be assumed that a high level of carbohydrates is necessary for growth. Alpha-glucosidases are important enzymes necessary for the ultimate liberation of glucose residues from various carbohydrates. Using acarbose affinity chromatog., a glycosylated alpha-glucosidase with a mol. mass of approx. 105 kDa was isolated for the first time from the hepatopancreas of the shrimp. Exhibiting an optimal catalytic activity in the temperature range from 40°C to 50°C at pH 6, the purified enzyme hydrolyzes α 1-4 bonds and liberates glucose from different oligo- and polysaccharides. By contrast to other known glucosidases, no α 1-6 glucose link with hydrolysis has been observed. This could explain the different rates of growth in shrimp aquaculture with starches from various origins. The amino-acid composition, together with the partial sequence of a hydrolytic peptide, shows a high degree of similarity to the alpha-glucosidases reported for various organisms including yeast and fungi and may help determine the phylogeny of the family.

REFERENCE COUNT: 50 THERE ARE 50 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 4 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1997:289431 CAPLUS

DOCUMENT NUMBER: 126:340302

TITLE: Efficient purification, characterization and partial amino acid sequencing of two α -1,4-glucan lyases from fungi

AUTHOR(S): Yu, Shukun; Christensen, Tove M. I. E.; Kragh, Karsten M.; Bojsen, Kirsten; Marcussen, Jan

CORPORATE SOURCE: Danisco Biotechnology, Danisco A/S, Langebrogade 1, PO box 17, DK 1001, Copenhagen K, Den.

SOURCE: Biochimica et Biophysica Acta, Protein Structure and Molecular Enzymology (1997), 1339(2), 311-320

CODEN: BBAEDZ; ISSN: 0167-4838

PUBLISHER: Elsevier B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB α -1,4-Glucan lyases from the fungi *Morchella costata* and *M. vulgaris* were purified by affinity chromatog. on β -cyclodextrin-sepharose, followed by ion exchange and gel filtration. The purified enzymes produced 1,5-anhydro-D-fructose from glucose oligomers and polymers with α -1,4-glucosidic linkages, such as maltose, maltosaccharides, amylopectin, and glycogen. The lyases were basically inactive towards glucans linked through α -1,1, α -1,3 or α -1,6 linkages. For both enzymes the mol. mass was around 121000 Da as determined by matrix-assisted laser desorption mass spectrometry. The pI for the lyases from *M. costata* and *M. vulgaris* was 4.5 and 4.4, resp. The lyases exhibited an optimal pH range of pH 5.5 to pH 7.5 with maximal activity at pH 6.5. Optimal temperature was between 37° and 48°

for the two lyases, depending on the substrates. The lyases were examined with 12 inhibitors to starch hydrolases and it was found that they were inhibited by the -SH group blocking agent PCMB and the following sugars and their analogs: glucose, maltitol, maltose, 1-deoxynojirimycin and acarbose. Partial amino acid sequences accounting for about 35% of the lyase polypeptides were determined. In the overlapping region of the sequences, the two lyases showed 91% identity. The two lyases also cross-reacted immunol.

REFERENCE COUNT: 25 THERE ARE 25 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 5 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1996:454496 CAPLUS

DOCUMENT NUMBER: 125:108807

TITLE: Thermostability of purified human pancreatic α -amylase is increased by the combination of Ca^{2+} and human serum albumin

AUTHOR(S): Tessier, Arthur J.; Dombi, Goerge W.; Bouwman, David L.

CORPORATE SOURCE: Dep. Surgery, Harper Hosp., Detroit, MI, 48201, USA

SOURCE: Clinica Chimica Acta (1996), 252(1), 11-20

CODEN: CCATAR; ISSN: 0009-8981

PUBLISHER: Elsevier

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Pancreatic fluid from a patient with a postoperative pancreatic fistula was used to isolate human α -amylase by means of acarbose affinity chromatog. Amylase thermostability was measured in 4 solns.: (1) EDTA-dialyzed; (2) dialyzed solution plus 0.15 mM (1.0 g/dL) human serum albumin; (3) dialyzed solution plus 0.25 mM (1.0 mg/dL) Ca^{2+} ; and (4) dialyzed solution with both human serum albumin and Ca^{2+} . Amylase activity was measured at predetd. times in samples heated to 60°. Thermostability was characterized by $t_{1/2}$, the time to 50% initial amylase enzyme activity. In the dialyzed solution $t_{1/2}$ was 0.75 min. This rose to 1.62 min with added human serum albumin, and to 8.24 min with added Ca^{2+} . The combination of human serum albumin and Ca^{2+} resulted in a synergistic increase of $t_{1/2}$ to 180 min. These findings support the authors' contention that human serum albumin, Ca^{2+} , and possibly other body fluid constituents must be considered in any utility involving amylase thermostability as a clin. relevant diagnostic marker.

L14 ANSWER 6 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1993:76027 CAPLUS

DOCUMENT NUMBER: 118:76027

TITLE: Interaction of catalytic-site mutants of *Bacillus subtilis* α -amylase with substrates and acarbose

AUTHOR(S): Takase, Kenji

CORPORATE SOURCE: Dep. Mol. Biol., Nat. Inst. Agrobiol. Resourc., Tsukuba, Japan

SOURCE: Biochimica et Biophysica Acta, Protein Structure and

Molecular Enzymology (1992), 1122(3), 278-82

CODEN: BBAEDZ; ISSN: 0167-4838

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The interactions of the 3 catalytic-site mutants of *B. subtilis* α -amylase/(D176N [Asp-176 \rightarrow Asn], E208Q [Glu-208 \rightarrow Gln], and D269N [Asp-269 \rightarrow Asn]) with substrates and a pseudooligosaccharide inhibitor, acarbose, were studied by means of difference absorption spectroscopy and affinity chromatog. The addition of maltopentaose or soluble starch to the inactive mutant enzymes mostly resulted in difference spectra characteristic of tryptophan perturbation, enabling the determination of the dissociation consts. The results showed that conversion of Glu-208 to Gln greatly enhanced substrate binding, implying that Glu-208 interacts

unfavorably with the substrate's ground state, preventing its optimal fit to the active site. The affinity for acarbose was greatly reduced in D269N and E208Q, but less so in D176N, suggesting that Asp-269 and Glu-208 are more important than Asp-176 in stabilizing the transition state. These results were consistent with Glu-208 and Asp-269 being the key catalytic residues, as previously proposed for Taka-amylase A.

L14 ANSWER 7 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1991:553644 CAPLUS
DOCUMENT NUMBER: 115:153644
TITLE: Characterization of thermostable α -glucosidase from *Clostridium thermohydrosulfuricum* 39E
AUTHOR(S): Saha, Badal C.; Zeikus, J. Gregory
CORPORATE SOURCE: Michigan Biotechnol. Inst., Lansing, MI, 48909, USA
SOURCE: Applied Microbiology and Biotechnology (1991), 35(5), 568-71
CODEN: AMBIDG; ISSN: 0175-7598
DOCUMENT TYPE: Journal
LANGUAGE: English

AB *C. thermohydrosulfuricum* 39E produced a cell-bound α -glucosidase. It was partially purified 140-fold by solubilizing with Triton X-100, (NH₄)₂SO₄ treatment, DEAE-Sepharose CL-6B, and octyl-Sepharose and acarbose-Sepharose affinity chromatog. The optimum temperature for enzyme activity was at 75°. It had a half-life of 35 min at 75°, 110 min at 70°, and 46 h at 60°. The enzyme was stable at pH 5.0-6.0 and had an optimum pH at 5.0-5.5. It hydrolyzed the A-1,4-linkages in maltose, maltotriose, maltotetraose, and maltohexaose, the rate decreasing in the order of higher-sized oligosaccharides. The enzyme preparation also hydrolyzed the A-1,6-linkages in isomaltose and isomaltotriose. It rapidly hydrolyzed p-nitrophenyl α -D-glucoside (pNPG). The K_m values for maltose, isomaltose, panose, maltotriose, and pNPG were 1.85, 2.95, 1.72, 0.58, and 0.31 mM, resp., at pH 5.5 and 60°. The enzyme produced glucose from all these substrates. The enzyme preparation did not require any metal ion for activity. The α -glucosidase activity was inhibited by acarbose.

L14 ANSWER 8 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1991:444768 CAPLUS
DOCUMENT NUMBER: 115:44768
TITLE: Topographical and enzymic characterization of amylases from the extremely thermophilic eubacterium *Thermotoga maritima*
AUTHOR(S): Schumann, Judith; Wrba, Alexander; Jaenicke, Rainer; Stetter, Karl Otto
CORPORATE SOURCE: Inst. Biophys. Phys. Biochem., Univ. Regensburg, Regensburg, D-8400, Germany
SOURCE: FEBS Letters (1991), 282(1), 122-6
CODEN: FEBLAL; ISSN: 0014-5793
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The hyperthermophilic eubacterium *T. maritima* uses starch as a substrate, without releasing amylase activity into the culture medium. The enzyme is associated with the toga. Its expression level is too low to allow the isolation of the pure enzyme. Using cycloheptaamylose and acarbose affinity chromatog. and common chromatog. procedures, two enzyme fractions are obtained. They differ in specificity, pH-optimum, temperature dependence and stability. Substrate specificity and Ca²⁺ dependence indicate α -, β - and gluco-amylase activity. Compared with α -amylase from *Bacillus licheniformis* (T_{max} = 75°), the amylases from *T. maritima* show exceedingly high thermal stability with an upper temperature limit at 95°. Significant turnover occurs only between 70 and 100°, i.e. in the range of viability of the microorganism.

L14 ANSWER 9 OF 17 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:209924 CAPLUS

DOCUMENT NUMBER: 106:209924

TITLE: Purification and characterization of extracellular α -amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678

AUTHOR(S): De Mot, Rene; Verachtert, Hubert

CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Heverlee, B-3030, Belg.

SOURCE: European Journal of Biochemistry (1987), 164(3), 643-54

CODEN: EJBCAI; ISSN: 0014-2956

DOCUMENT TYPE: Journal

LANGUAGE: English

AB An α -amylase and a glucoamylase were purified to homogeneity from the culture fluid of β -cyclodextrin-grown *C. antarctica* CBS 6678 by protamine sulfate treatment, $(\text{NH}_4)_2\text{SO}_4$ precipitation, gel filtration (Sephadex G-75 sf, Ultrogel ACA 54), DEAE-Sephacel chromatog., hydroxyapatite chromatog., and affinity chromatog. on acarbose-AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compns. Their apparent relative mol. mass, sedimentation coefficient ($s_{20,w}^\circ$), pI, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7

S, 10.1,

1.74 $\text{cm}^2 \text{mg}^{-1}$, 4.2°, and 57°, resp., for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 $\text{cm}^2 \text{mg}^{-1}$, 4.2° and 62°, resp., for α -amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-mol.-mass substrates, including some raw starches. The α -amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both α -amylase ($K_i < 1 \mu\text{M}$) and glucoamylase ($K_i < 0.1 \mu\text{M}$), being more effective than Bay e 4609 ($K_i < 10 \mu\text{M}$). Glucoamylase was selectively and strongly inhibited by acarbose ($K_i < 0.1 \mu\text{M}$). Activity of the latter enzyme was also affected by 1-deoxynojirimycin ($K_i < 1 \text{mM}$), maltitol, and amino alcs. ($K_i < 10 \text{mM}$). Unlike α -amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being nonidentical with the active site.

L14 ANSWER 10 OF 17 MEDLINE on STN

ACCESSION NUMBER: 2005550205 MEDLINE

DOCUMENT NUMBER: PubMed ID: 16198511

TITLE: Enzymatic characterization of a maltogenic amylase from *Lactobacillus gasser* ATCC 33323 expressed in *Escherichia coli*.

AUTHOR: Oh Ko-Woon; Kim Myo-Jeong; Kim Hae-Yeong; Kim Byung-Yong; Baik Moo-Yeol; Auh Joong-Hyuck; Park Cheon-Seok

CORPORATE SOURCE: Department of Food Science and Biotechnology, Institute of Life Sciences and Resources, KyungHee University, Yongin 449-701, South Korea.

SOURCE: FEMS microbiology letters, (2005 Nov 1) Vol. 252, No. 1, pp. 175-81. Electronic Publication: 2005-09-19. Journal code: 7705721. ISSN: 0378-1097.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200512

ENTRY DATE: Entered STN: 18 Oct 2005

Last Updated on STN: 18 Dec 2005

Entered Medline: 12 Dec 2005

AB A gene corresponding to a maltogenic amylase (MAase) in *Lactobacillus gasser* ATCC 33323 (lgma) was cloned and expressed in *Escherichia coli*. The recombinant LGMA was efficiently purified 24.3-fold by one-step Ni-NTA

affinity chromatography. The final yield and specific activity of the purified recombinant LGMA were 68% and 58.7 U/mg, respectively. The purified enzyme exhibited optimal activity for beta-CD hydrolysis at 55 degrees C and pH 5. The relative hydrolytic activities of LGMA to beta-CD, soluble starch or pullulan was 8:1:1.9. The activity of LGMA was strongly inhibited by most metal ions, especially Zn(2+), Fe(2+), Co(2+) and by EDTA. LGMA possessed some unusual properties distinguishable from typical MAases, such as being in a tetrameric form, having hydrolyzing activity towards the alpha-(1,6)-glycosidic linkage and being inhibited by acarbose.

L14 ANSWER 11 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 2003358724 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 12890998
 TITLE: Inhibitory effects of human and porcine alpha-amylase on CCK-8-stimulated lipase secretion of isolated rat pancreatic acini.
 AUTHOR: Jonas Ludwig; Mikkat Ulrike; Lehmann Renate; Schareck Wolfgang; Walzel Hermann; Schroder Werner; Lopp Hilja; Pussa Tonu; Toomik Peeter
 CORPORATE SOURCE: Department of Pathology, Faculty of Medicine, University of Rostock, Germany.. ludwig.jonas@med.uni-rostock.de
 SOURCE: Pancreatology : official journal of the International Association of Pancreatology (IAP) ... [et al.], (2003) Vol. 3, No. 4, pp. 342-8.
 Journal code: 100966936. ISSN: 1424-3903.
 PUB. COUNTRY: Switzerland
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 200403
 ENTRY DATE: Entered STN: 1 Aug 2003
 Last Updated on STN: 17 Mar 2004
 Entered Medline: 16 Mar 2004
 AB Previously we have demonstrated inhibitory effects of the plant lectin wheat germ agglutinin (WGA) on (125)I-CCK-8 binding to pancreatic AR42J cells as well as on CCK-8-stimulated Ca(2+) release and alpha-amylase secretion of rat pancreatic acini or acinar cells. Therefore, it is entirely conceivable that alpha-amylase having several lectin-like carbohydrate recognition domains can modulate the CCK-8 stimulated lipase secretion. Human alpha-amylase, purified from pancreatic juice by affinity chromatography to homogeneity, and commercial porcine pancreatic alpha-amylase inhibit CCK-8-stimulated lipase secretion of rat pancreatic acini in a concentration-dependent manner. Acarbose, a specific inhibitor of alpha-amylase, was without effect on CCK-8-induced cellular lipase secretion. The data presented here provide evidence for a regulatory function of alpha-amylase in CCK-8-stimulated pancreatic secretion.
 Copyright 2003 S. Karger AG, Basel and IAP

L14 ANSWER 12 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 1998203259 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 9542155
 TITLE: alpha-Glucosidase from the hepatopancreas of the shrimp, Penaeus vannamei (Crustacea-Decapoda).
 AUTHOR: Le Chevalier P; Van Wormhoudt A
 CORPORATE SOURCE: Institut Universitaire de Technologie, Quimper, France.. chevalie@iutquimp.univ-brest.fr
 SOURCE: The Journal of experimental zoology, (1998 Apr 15) Vol. 280, No. 6, pp. 384-94.
 Journal code: 0375365. ISSN: 0022-104X.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English

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(FILE 'HOME' ENTERED AT 16:13:05 ON 06 SEP 2006)

FILE 'CAPLUS, MEDLINE' ENTERED AT 16:13:24 ON 06 SEP 2006

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L2      5 S ACARBOSE (P) ALCOHOL? (P) ENZYME?
L3      2 S L2 NOT L1
L4      3 S ACARBOSE (P) ALCOHOL? (P) CHROMATOGRAPHY
L5      1 S ACARBOSE (P) ETHANOL? (P) CHROMATOGRAPHY
L6      5 S ACARBOSE (P) ETHANOL? (P) ENZYME?
L7      3 S ACARBOSE (P) PURIFICATION (P) ENZYME (P) AFFINITY
L8      3 S ACARBOSE (P) PURIFICATION (P) FERMENTATION BROTH?
L9     14 S ACARBOSE (P) PURIFICATION (P) CHROMATOGRAPHY
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L12    25 S ACARBOSE (P) AFFINITY CHROMATOGRAPHY
L13    26084 S L12 NOT L9
L14    17 S L12 NOT L9
L15     0 S ACARBOSE (P) ETHYL ALCOHOL? CHROMATOGRAPHY
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L17     0 S ACARBOSE (P) ETHYL ALCOHOL? (P) PURE
L18     0 S ACARBOSE (P) ETHYL ALCOHOL? (P) PURIFY
L19     0 S ACARBOSE (P) ETHYL ALCOHOL? (P) PURIFICATION
L20     3 S ACARBOSE (P) ETHANOL? (P) PURIFICATION
L21    47 S ACARBOSE (P) ENZYME (P) CHROMATOGRAPHY
L22     1 S ACARBOSE (P) IMMOBIL? ENZYME (P) CHROMATOGRAPHY
L23     0 S ACARBOSE (P) ENZYME CHROMATOGRAPHY
L24    10 S ACARBOSE (P) ENZYME (P) SUPPORT
L25   112 S ACARBOSE (P) ?GLUCOAMYLASE
L26    18 S L25 AND PURIFICATION?
L27     1 S L25 AND PURE
L28     8 S L25 AND COLUMN
L29    20 S L25 AND CHROMATOGRAPHY
L30    14 S ACARBOSE/TI AND CHROMATOGRAPH?/TI
L31     0 S ACARBOSE (P) AMMONIA (P) CHRMATOGRAPHY
L32     3 S ACARBOSE (P) ETHANOL (P) PURIFICATION
L33     4 S ACARBOSE (P) ETHANOL (P) PUR?
L34     1 S ACARBOSE (P) AMMONIA (P) PURIFICATION
L35     1 S ACARBOSE (P) AMMONIA (P) PUR?
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FILE SEGMENT: Priority Journals
ENTRY MONTH: 199805
ENTRY DATE: Entered STN: 20 May 1998
Last Updated on STN: 18 Dec 2002
Entered Medline: 8 May 1998

AB *Penaeus vannamei* is an omnivorous species, and it can be assumed that a high level of carbohydrates is necessary for growth. Alpha-glucosidases are important enzymes necessary for the ultimate liberation of glucose residues from various carbohydrates. Using acarbose affinity chromatography, a glycosylated alpha-glucosidase with a molecular mass of approximately 105 kDa was isolated for the first time from the hepatopancreas of the shrimp. Exhibiting an optimal catalytic activity in the temperature range from 40 degrees C to 50 degrees C at pH 6, the purified enzyme hydrolyses alpha 1-4 bonds and liberates glucose from different oligo and polysaccharides. By contrast to other known glucosidases, no alpha 1-6 glucose link with hydrolysis has been observed. This could explain the different rates of growth in shrimp aquaculture with starches from various origins. The amino-acid composition, together with the partial sequence of a hydrolytic peptide, shows a high degree of similarity to the alpha-glucosidases reported for various organisms including yeast and fungi and may help determine the phylogeny of the family.

L14 ANSWER 13 OF 17 MEDLINE on STN
ACCESSION NUMBER: 97330817 MEDLINE
DOCUMENT NUMBER: PubMed ID: 9187252
TITLE: Efficient purification, characterization and partial amino acid sequencing of two alpha-1,4-glucan lyases from fungi.
AUTHOR: Yu S; Christensen T M; Kragh K M; Bojsen K; Marcussen J
CORPORATE SOURCE: Danisco Biotechnology, Danisco A/S, Langebrogade 1, Copenhagen K, Denmark.. g7sy@danisco.dk
SOURCE: Biochimica et biophysica acta, (1997 May 23) Vol. 1339, No. 2, pp. 311-20.
Journal code: 0217513. ISSN: 0006-3002.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199707
ENTRY DATE: Entered STN: 16 Jul 1997
Last Updated on STN: 16 Jul 1997
Entered Medline: 1 Jul 1997

AB alpha-1,4-Glucan lyases from the fungi *Morchella costata* and *M. vulgaris* were purified by affinity chromatography on beta-cyclodextrin-sepharose, followed by ion exchange and gel filtration. The purified enzymes produced 1,5-anhydro-D-fructose from glucose oligomers and polymers with alpha-1,4-glucosidic linkages, such as maltose, maltosaccharides, amylopectin, and glycogen. The lyases were basically inactive towards glucans linked through alpha-1,1, alpha-1,3 or alpha-1,6 linkages. For both enzymes the molecular mass was around 121,000 Da as determined by matrix-assisted laser desorption mass spectrometry. The pI for the lyases from *M. costata* and *M. vulgaris* was 4.5 and 4.4, respectively. The lyases exhibited an optimal pH range of pH 5.5 to pH 7.5 with maximal activity at pH 6.5. Optimal temperature was between 37 degrees C and 48 degrees C for the two lyases, depending on the substrates. The lyases were examined with 12 inhibitors to starch hydrolases and it was found that they were inhibited by the -SH group blocking agent PCMB and the following sugars and their analogues: glucose, maltitol, maltose, 1-deoxynojirimycin and acarbose. Partial amino acid sequences accounting for about 35% of the lyase polypeptides were determined. In the overlapping region of the sequences, the two lyases showed 91% identity. The two lyases also cross-reacted immunologically.

L14 ANSWER 14 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 96409375 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 8814357
 TITLE: Thermostability of purified human pancreatic alpha-amylase is increased by the combination of Ca²⁺ and human serum albumin.
 AUTHOR: Tessier A J; Dombi G W; Bouwman D L
 CORPORATE SOURCE: Harper Hospital, Department of Surgery, Detroit, MI 48201, USA. atessie/cms.cc.wayne.edu.
 SOURCE: Clinica chimica acta; international journal of clinical chemistry, (1996 Aug 15) Vol. 252, No. 1, pp. 11-20. Journal code: 1302422. ISSN: 0009-8981.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199612
 ENTRY DATE: Entered STN: 28 Jan 1997
 Last Updated on STN: 28 Jan 1997
 Entered Medline: 18 Dec 1996

AB Pancreatic fluid from a patient with a post operative pancreatic fistula was used to isolate human alpha-amylase by means of acarbose affinity chromatography. Amylase thermostability was measured in 4 solutions: (1) EDTA-dialyzed; (2) dialyzed solution plus 0.15 mmol/l (1.0 g/dl) human serum albumin; (3) dialyzed solution plus 0.25 mmol/l (1.0 mg/dl) calcium ions; and (4) dialyzed solution with both human serum albumin and calcium ions. Amylase activity was measured at predetermined times in samples heated to 60 degrees C. Thermostability was characterized by t_{1/2}, the time to 50% initial amylase enzyme activity. In the dialyzed solution t_{1/2} was 0.75 +/- 0.19 min. This rose to 1.62 +/- 0.34 min with added human serum albumin, and to 8.24 +/- 0.13 min with added calcium ions. The combination of human serum albumin and calcium ions resulted in a synergistic increase of t_{1/2} to 180 +/- 26 min. These findings support our contention that human serum albumin, calcium ions and possibly other body fluid constituents must be considered in any utility involving amylase thermostability as a clinically relevant diagnostic marker.

L14 ANSWER 15 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 92369111 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 1380303
 TITLE: Interaction of catalytic-site mutants of Bacillus subtilis alpha-amylase with substrates and acarbose.
 AUTHOR: Takase K
 CORPORATE SOURCE: Department of Molecular Biology, National Institute of Agrobiological Resources, Ibaraki, Japan.
 SOURCE: Biochimica et biophysica acta, (1992 Aug 21) Vol. 1122, No. 3, pp. 278-82. Journal code: 0217513. ISSN: 0006-3002.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199209
 ENTRY DATE: Entered STN: 9 Oct 1992
 Last Updated on STN: 3 Mar 2000
 Entered Medline: 22 Sep 1992

AB The interactions of the three catalytic-site mutants of Bacillus subtilis alpha-amylase/(DN176 [Asp-176----Asn], EQ208 [Glu-208----Gln] and DN269 [Asp-269----Asn]) with substrates and a pseudo-oligosaccharide inhibitor, acarbose, have been studied by means of difference absorption spectroscopy and affinity chromatography. The addition of maltopentaose or soluble starch to the inactive mutant enzymes mostly resulted in difference spectra characteristic of tryptophan

perturbation, enabling determination of the dissociation constants. The results show that conversion of Glu-208 to Gln greatly enhanced substrate binding, implying that Glu-208 interacts unfavorably with the substrate's ground state, preventing its optimal fit to the active site. The affinity for acarbose was greatly reduced in DN269 and EQ208, but less so in DN176, suggesting that Asp-269 and Glu-208 are more important than Asp-176 in stabilizing the transition state. These results are consistent with Glu-208 and Asp-269 being the key catalytic residues, as proposed for Taka-amylase A.

L14 ANSWER 16 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 91224312 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 1709115
 TITLE: Topographical and enzymatic characterization of amylases from the extremely thermophilic eubacterium *Thermotoga maritima*.
 AUTHOR: Schumann J; Wrba A; Jaenicke R; Stetter K O
 CORPORATE SOURCE: Institut fur Biophysik und Physikalische Biochemie, Universitat Regensburg, Germany.
 SOURCE: FEBS letters, (1991 Apr 22) Vol. 282, No. 1, pp. 122-6. Journal code: 0155157. ISSN: 0014-5793.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199106
 ENTRY DATE: Entered STN: 30 Jun 1991
 Last Updated on STN: 29 Jan 1996
 Entered Medline: 12 Jun 1991

AB The hyperthermophilic eubacterium *Thermotoga maritima* uses starch as a substrate, without releasing amylase activity into the culture medium. The enzyme is associated with the 'toga'. Its expression level is too low to allow the isolation of the pure enzyme. Using cycloheptaamylose and acarbose affinity chromatography and common chromatographic procedures, two enzyme fractions are obtained. They differ in specificity, pH-optimum, temperature dependence and stability. Substrate specificity and Ca²⁺ dependence indicate alpha-, beta- and gluco-amylase activity. Compared with alpha-amylase from *Bacillus licheniformis* (T_{max} = 75 degrees C), the amylases from *Thermotoga maritima* show exceedingly high thermal stability with an upper temperature limit at 95 degrees C. Significant turnover occurs only between 70 and 100 degrees C, i.e. in the range of viability of the microorganism.

L14 ANSWER 17 OF 17 MEDLINE on STN
 ACCESSION NUMBER: 87190439 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 3106037
 TITLE: Purification and characterization of extracellular alpha-amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678.
 AUTHOR: De Mot R; Verachtert H
 SOURCE: European journal of biochemistry / FEBS, (1987 May 4) Vol. 164, No. 3, pp. 643-54. Journal code: 0107600. ISSN: 0014-2956.
 PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198706
 ENTRY DATE: Entered STN: 3 Mar 1990
 Last Updated on STN: 3 Mar 1990
 Entered Medline: 25 Jun 1987

AB An alpha-amylase and a glucoamylase were purified to homogeneity from the culture fluid of beta-cyclodextrin-grown *Candida antarctica* CBS 6678 by protamine sulfate treatment, ammonium sulfate precipitation, gel

filtration (Sephadex G-75 sf, Ultrogel ACA 54), DEAE-Sephacel chromatography, hydroxyapatite chromatography and affinity chromatography on acarbose--AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compositions. Their apparent relative molecular mass, sedimentation coefficient ($S_{20,w}$), isoelectric point, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2 and 57 degrees C, respectively, for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2 and 62 degrees C, respectively, for alpha-amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-molecular-mass substrates, including some raw starches. alpha-Amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both alpha-amylase (K_i less than 1 microM) and glucoamylase (K_i less than 0.1 microM), being more effective than Bay e 4609 (K_i less than 10 microM). Glucoamylase was selectivity and strongly inhibited by acarbose (K_i less than 0.1 microM). Activity of the latter enzyme was also affected by 1-deoxynojirimycin (K_i less than 1 mM), maltitol and amino alcohols (K_i less than 10 mM). Unlike alpha-amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being non-identical with the active site.

L20 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purification process for manufacturing a high pure acarbose relates to a process for preparing high pure acarbose from acarbose-containing fermentation broth. The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L20 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:928233 CAPLUS
DOCUMENT NUMBER: 138:3755
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Hung.
SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S. Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|------|----------|-----------------|----------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-223492P P 20000807
US 2001-924271 A2 20010807

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing

the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of a sodium chloride solution and a salt solution; precipitating the acarbose with a solvent; and recovering the precipitated acarbose.

L20 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN
 ACCESSION NUMBER: 2002:123021 CAPLUS
 DOCUMENT NUMBER: 136:182542
 TITLE: Method for purification of acarbose
 INVENTOR(S): Keri, Vilmos; Deak, Lajos
 PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.
 SOURCE: PCT Int. Appl., 24 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 2
 PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|------------|
| WO 2002012256 | A1 | 20020214 | WO 2001-US24729 | 20010807 |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
| AU 2001084741 | A5 | 20020218 | AU 2001-84741 | 20010807 |
| EP 1309601 | A1 | 20030514 | EP 2001-963821 | 20010807 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | WO 2001-US24729 | W 20010807 |

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of hydrochloric acid and the weak acid; precipitating the acarbose with a solvent; and separating the acarbose crystals.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L26 ANSWER 1 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2006:661962 CAPLUS
TITLE: Inhibition of recombinant human maltase glucoamylase
by salacinol and derivatives
AUTHOR(S): Rossi, Elena J.; Sim, Lyann; Kuntz, Douglas A.; Hahn,
Dagmar; Johnston, Blair D.; Ghavami, Ahmad; Szczepina,
Monica G.; Kumar, Nag S.; Sterchi, Erwin E.; Nichols,
Buford L.; Pinto, B. M.; Rose, David R.
CORPORATE SOURCE: Department of Medical Biophysics, University of
Toronto, Can.
SOURCE: FEBS Journal (2006), 273(12), 2673-2683
CODEN: FJEOAC; ISSN: 1742-464X
PUBLISHER: Blackwell Publishing Ltd.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Inhibitors targeting pancreatic α -amylase and intestinal
 α -glucosidases delay glucose production following digestion and are
currently used in the treatment of Type II diabetes. Maltase-
glucoamylase (MGA), a family 31 glycoside hydrolase, is an
 α -glucosidase anchored in the membrane of small intestinal
epithelial cells responsible for the final step of mammalian starch
digestion leading to the release of glucose. This paper reports the
production and purifn. of active human recombinant MGA amino
terminal catalytic domain (MGAnt) from two different eukaryotic cell
culture systems. MGAnt overexpressed in Drosophila cells was of quality
and quantity suitable for kinetic and inhibition studies as well as future
structural studies. Inhibition of MGAnt was tested with a group of
prospective α -glucosidase inhibitors modeled after salacinol, a
naturally occurring α -glucosidase inhibitor, and acarbose,
a currently prescribed antidiabetic agent. Four synthetic inhibitors that
bind and inhibit MGAnt activity better than acarbose, and at
comparable levels to salacinol, were found. The inhibitors are derivs. of
salacinol that contain either a selenium atom in place of sulfur in the
five-membered ring, or a longer polyhydroxylated, sulfated chain than
salacinol. Six-membered ring derivs. of salacinol and compds. modeled
after miglitol were much less effective as MGAnt inhibitors. These
results provide information on the inhibitory profile of MGAnt that will
guide the development of new compds. having antidiabetic activity.

REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L26 ANSWER 2 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a
high purity acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|------|----------|-----------------|----------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |

PRIORITY APPLN. INFO.: TW 2003-92133913 A 20031202

AB A purifn. process for manufacturing a high pure acarbose relates to a
process for preparing high pure acarbose from acarbose-containing fermentation
broth.

The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L26 ANSWER 3 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2003:356465 CAPLUS
DOCUMENT NUMBER: 138:334026
TITLE: High throughput isolation of biological compounds
INVENTOR(S): Frisner, Henrik; Christensen, Lars Lehmann Hylling
PATENT ASSIGNEE(S): Novozymes A/S, Den.
SOURCE: PCT Int. Appl., 37 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|------------|
| ----- | ---- | ----- | ----- | ----- |
| WO 2003037914 | A2 | 20030508 | WO 2002-DK717 | 20021029 |
| WO 2003037914 | A3 | 20040325 | | |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
| CA 2465543 | AA | 20030508 | CA 2002-2465543 | 20021029 |
| AU 2002340776 | A1 | 20030512 | AU 2002-340776 | 20021029 |
| EP 1442048 | A2 | 20040804 | EP 2002-774472 | 20021029 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK | | | | |
| PRIORITY APPLN. INFO.: | | | DK 2001-1598 | A 20011030 |
| | | | WO 2002-DK717 | W 20021029 |

AB A method for high throughput micro-purifn. of a library of tag-free biol. compds. is claimed wherein a population of discrete liquid samples comprising the library of tag-free biol. compds. is contacted with a solid chromatog. material comprising a functional group selected from ion exchange materials, hydrophobic materials, affinity materials and hydrophobic charge induction materials so as to retain the tag-free biol. compds. of the library, isolating the chromatog. material retaining the biol. compds., releasing the biol. compds. of the library from the solid chromatog. material and collecting the released biol. compds. of the library to produce a population of samples comprising the isolated biol. compound

L26 ANSWER 4 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1994:100047 CAPLUS
DOCUMENT NUMBER: 120:100047
TITLE: Purification and characterization of a pullulan-hydrolyzing glucoamylase from Sclerotium rolfsii
AUTHOR(S): Kelkar, Hemant S.; Deshpande, Mukund V.
CORPORATE SOURCE: Pune, India
SOURCE: Starch/Staerke (1993), 45(10), 361-8
CODEN: STARDD; ISSN: 0038-9056
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The pullulan-hydrolyzing enzyme from culture filtrates of Sclerotium

rolfsii grown on soluble starch as a carbon source has been purified by ultrafiltration (Amicon, PM-10), ion-exchange chromatog. (DEAE-Cellulose DE-52) and gel filtration chromatog. (Bio-Gel P-150). The enzyme moved as a single band in non-denaturing polyacrylamide gel electrophoresis carried out at pH 2.9 and 7.5. The relative mol. mass of the enzyme was estimated to be 64,000 D by SDS-PAGE and 66,070 D by gel filtration on Bio-Gel P150. The enzyme hydrolyzed pullulan optimally at 50°C between pH 4.0-4.5, whereas, soluble starch was optimally hydrolyzed at a pH of between 4.0-4.5 and at 65°C. The Michaelis constant (Km) for pullulan was 5.13 mg·mL⁻¹ (Vmax 1.0 U·mg⁻¹) and for soluble starch, it was 0.6 mg·mL⁻¹ (Vmax 8.33 U·mg⁻¹). The enzyme was observed to be a glycoprotein (12-13% carbohydrate by weight) and had a strong affinity for Con A. The enzyme hydrolyzed α-D-glucans in an exo-manner, which resulted in the release of glucose as the sole product of hydrolysis. Acarbose, a maltotetraose analog, was found to be a potent inhibitor of both pullulan and starch hydrolysis (100% inhibition at 0.06 μM). The enzyme has been characterized as a glucoamylase (1,4-α-D-glucan glucohydrolase, EC 3.2.1.3) showing a significant action on pullulan.

L26 ANSWER 5 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:524834 CAPLUS

DOCUMENT NUMBER: 109:124834

TITLE: Effective purification of glucoamylase in koji, a solid culture of *Aspergillus oryzae* on steamed rice, by affinity chromatography using an immobilized acarbose (BAY g-5421)

AUTHOR(S): Ono, Kazuhisa; Shigeta, Seiko; Oka, Satoru

CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan

SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1707-14

CODEN: ABCHA6; ISSN: 0002-1369

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Glucoamylase (GA) was purified from koji, a solid culture of *A. oryzae* on steamed rice, by extraction with 1% NaCl solution, precipitation with EtOH, and acarbose affinity chromatog. The purified enzyme was homogeneous on gel filtration, PAGE and SDS-PAGE, ultracentrifugation, and IEF. The enzyme released β-glucose as a sole product from soluble starch and maltooligosaccharides. The other common and inherent features of GAs were also confirmed in the GA from *A. oryzae*. The enzyme was a glycoprotein containing .apprx.4.8% glucosamine and 7.8% neutral saccharides.

L26 ANSWER 6 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:419287 CAPLUS

DOCUMENT NUMBER: 109:19287

TITLE: Purification and characterization of a highly thermostable novel pullulanase from *Clostridium thermohydrosulfuricum*

AUTHOR(S): Saha, Badal C.; Mathupala, Saroj P.; Zeikus, J. Gregory

CORPORATE SOURCE: Michigan Biotechnol. Inst., Lansing, MI, 48909, USA

SOURCE: Biochemical Journal (1988), 252(2), 343-8

CODEN: BIJOAK; ISSN: 0306-3275

DOCUMENT TYPE: Journal

LANGUAGE: English

AB *C. thermohydrosulfuricum* Mutant Z 21-109 produced intracellular thermostable pullulanase and glucoamylase activities. The glucoamylase activity was inactivated by treating *C. thermohydrosulfuricum* cells with 10% propan-1-ol at 85° in the presence of 5 mM CaCl₂. Pullulanase activity was selectively solubilized

from cells by treatment with detergent and lipase. The solubilized pullulanase was purified by treatment with streptomycin sulfate and $(\text{NH}_4)_2\text{SO}_4$ and by DEAE-Sephacel, octyl-Sepharose, and pullulan-Sepharose chromatog. Pullulanase was purified 3511-fold and displayed homogeneity on SDS-PAGE. The pullulanase was a monomeric glycoprotein with an apparent mol. weight of .apprx.136,500, and it displayed a pI of 5.9. The enzyme was enriched in both acidic and hydrophobic amino acids. The purified pullulanase was stable and optimally active at 90°. The optimum pH for activity and pH-stability ranges were 5.0-5.5 and 3.0-5.0, resp. The enzyme was inhibited by cyclodextrins, EDTA, and N-bromosuccinimide, but not by p-chloromercuribenzoate and acarbose. Pullulanase displayed a relative substrate specificity for hydrolysis of pullulan (100%) vs. 75% for glycogen and 50% for soluble starch. The apparent K_m , V_{max} , and catalytic rate constant values for enzyme activity on pullulan at 60° were 0.675 mg/mL, 122.5 μmol of reducing sugar formed/min/mg of protein, and 16,240 min^{-1} resp. The novel properties of this extremely thermostable pullulanase are discussed in relation to other purified starch-debranching enzymes.

L26 ANSWER 7 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:209924 CAPLUS

DOCUMENT NUMBER: 106:209924

TITLE: Purification and characterization of extracellular α -amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678

AUTHOR(S): De Mot, Rene; Verachtert, Hubert

CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Heverlee, B-3030, Belg.

SOURCE: European Journal of Biochemistry (1987), 164(3), 643-54

CODEN: EJBCAI; ISSN: 0014-2956

DOCUMENT TYPE: Journal

LANGUAGE: English

AB An α -amylase and a glucoamylase were purified to homogeneity from the culture fluid of β -cyclodextrin-grown *C. antarctica* CBS 6678 by protamine sulfate treatment, $(\text{NH}_4)_2\text{SO}_4$ precipitation, gel

filtration (Sephadex G-75 sf, Ultrogel AcA 54), DEAE-Sephacel chromatog., hydroxyapatite chromatog., and affinity chromatog. on acarbose -AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compns. Their apparent relative mol. mass, sedimentation coefficient ($s_{20,w}$), pI, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 $\text{cm}^2 \text{mg}^{-1}$, 4.2°, and 57°, resp., for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 $\text{cm}^2 \text{mg}^{-1}$, 4.2° and 62°, resp., for α -amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-mol.-mass substrates, including some raw starches. The α -amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both α -amylase ($K_i < 1 \mu\text{M}$) and glucoamylase ($K_i < 0.1 \mu\text{M}$), being more effective than Bay e 4609 ($K_i < 10 \mu\text{M}$). Glucoamylase was selectively and strongly inhibited by acarbose ($K_i < 0.1 \mu\text{M}$). Activity of the latter enzyme was also affected by 1-deoxynojirimycin ($K_i < 1 \text{ mM}$), maltitol, and amino alcs. ($K_i < 10 \text{ mM}$). Unlike α -amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being nonidentical with the active site.

L26 ANSWER 8 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS

DOCUMENT NUMBER: 105:167504

TITLE: Purification of glucoamylase by acarbose (BAY g-5421) affinity chromatography

AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.

CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA
SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3),
201-9
CODEN: BABIEC; ISSN: 0885-4513
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was purified from com. available, impure enzyme prepns. by affinity chromatog. on acarbose (II) columns. Up to 2 mg I was bound without leakage to a 1-mL affinity gel column possessing a covalently linked II ligand (1 mg II/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purifn. was accomplished in .apprx.8 h. Both I activities were recovered in >80% yield, free of α -amylase activity and possessing specific activities comparable to those of prepns. obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, II affinity chromatog. provides a general method for the rapid and efficient purifn. of I, and appears to be ideally suited for scale-up for the com. purifn. of these enzymes.

L26 ANSWER 9 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1986:125516 CAPLUS
DOCUMENT NUMBER: 104:125516
TITLE: Purification and characterization of an extracellular glucoamylase from the yeast *Candida tsukubaensis* CBS 6389
AUTHOR(S): De Mot, R.; Van Oudendijck, E.; Verachtert, H.
CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Louvain, B-3030, Belg.
SOURCE: Antonie van Leeuwenhoek (1985), 51(3), 275-87
CODEN: ALJMAO; ISSN: 0003-6072
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The starch-degrading yeast *C. tsukubaensis* CBS 6389 secreted amylase with high activity when grown in a medium containing soluble starch. The extracellular α -amylase activity was very low. The major amylase component was purified by DEAE-Sephadex A-50 chromatog. and Ultrogel Aca 44 gel filtration and characterized as a glucoamylase. The enzyme was a glycoprotein with a mol. weight of 56,000. The glucoamylase had a temperature optimum of 55°, and its highest activity was at pH 2.4-4.8. Acarbose strongly inhibited the purified glucoamylase. Debranching activity was present, as demonstrated by the release of glucose from pullulan.

L26 ANSWER 10 OF 18 CAPLUS COPYRIGHT 2006 ACS on STN
ACCESSION NUMBER: 1986:30774 CAPLUS
DOCUMENT NUMBER: 104:30774
TITLE: Purification and characterization of extracellular amylolytic enzymes from the yeast *Filobasidium capsuligenum*
AUTHOR(S): De Mot, Rene; Verachtert, Hubert
CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Louvain, B-3030, Belg.
SOURCE: Applied and Environmental Microbiology (1985), 50(6), 1474-82
CODEN: AEMIDF; ISSN: 0099-2240
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The extracellular amylolytic system of *Filobasidium capsuligenum* consisted of an α -amylase (EC 3.2.1.1) (I) and 2 forms of glucoamylase (EC 3.2.1.3) (II). The enzymes were purified by (NH₄)₂SO₄ fractionation, repeated ion-exchange chromatog. (DEAE-Sephadex A-50), and gel filtration (Sephadex G-25, Sephadex G-100). I had an optimum pH of 5.6 and an

optimum temperature of 50°, but was rapidly inactivated at higher temperature. The mol. weight was estimated by SDS-polyacrylamide gel electrophoresis to be 64,000. An acarbose concentration of 20 µg/mL was required for 50% inhibition of I. Both II enzymes were glycoproteins of identical mol. weight (60,000) and produced only glucose by exohydrolysis. The debranching activity of the II enzymes was evidenced with substrates containing α-1,6 linkages. The pH optima were 5.0-5.6 for II-I and 4.8-5.3 for II-II. II-I had a higher optimum temperature (55%) than II-II (50%) and was also more resistant to thermal inactivation. Only low acarbose concns. (<0.1 µg/mL) were required to reduce the activity of II-I and II-II by 50%.

L26 ANSWER 11 OF 18 MEDLINE on STN
 ACCESSION NUMBER: 2006399279 IN-PROCESS
 DOCUMENT NUMBER: PubMed ID: 16817895
 TITLE: Inhibition of recombinant human maltase glucoamylase by salacinol and derivatives.
 AUTHOR: Rossi Elena J; Sim Lyann; Kuntz Douglas A; Hahn Dagmar; Johnston Blair D; Ghavami Ahmad; Szczepina Monica G; Kumar Nag S; Sterchi Erwin E; Nichols Buford L; Pinto B M; Rose David R
 CORPORATE SOURCE: Department of Medical Biophysics, University of Toronto, Canada.
 SOURCE: The FEBS journal, (2006 Jun) Vol. 273, No. 12, pp. 2673-83. Journal code: 101229646. ISSN: 1742-464X.
 PUB. COUNTRY: England: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: NONMEDLINE; IN-PROCESS; NONINDEXED; Priority Journals
 ENTRY DATE: Entered STN: 6 Jul 2006
 Last Updated on STN: 14 Jul 2006

AB Inhibitors targeting pancreatic alpha-amylase and intestinal alpha-glucosidases delay glucose production following digestion and are currently used in the treatment of Type II diabetes. Maltase-glucoamylase (MGA), a family 31 glycoside hydrolase, is an alpha-glucosidase anchored in the membrane of small intestinal epithelial cells responsible for the final step of mammalian starch digestion leading to the release of glucose. This paper reports the production and purification of active human recombinant MGA amino terminal catalytic domain (MGAnt) from two different eukaryotic cell culture systems. MGAnt overexpressed in Drosophila cells was of quality and quantity suitable for kinetic and inhibition studies as well as future structural studies. Inhibition of MGAnt was tested with a group of prospective alpha-glucosidase inhibitors modeled after salacinol, a naturally occurring alpha-glucosidase inhibitor, and acarbose, a currently prescribed antidiabetic agent. Four synthetic inhibitors that bind and inhibit MGAnt activity better than acarbose, and at comparable levels to salacinol, were found. The inhibitors are derivatives of salacinol that contain either a selenium atom in place of sulfur in the five-membered ring, or a longer polyhydroxylated, sulfated chain than salacinol. Six-membered ring derivatives of salacinol and compounds modeled after miglitol were much less effective as MGAnt inhibitors. These results provide information on the inhibitory profile of MGAnt that will guide the development of new compounds having antidiabetic activity.

L26 ANSWER 12 OF 18 MEDLINE on STN
 ACCESSION NUMBER: 2006040708 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 16182318
 TITLE: Insulin sensitizing and alpha-glucoamylase inhibitory action of sennosides, rheins and rhaponticin in Rhei Rhizoma.
 AUTHOR: Choi Soo Bong; Ko Byoung Seob; Park Seong Kyu; Jang Jin Sun; Park Sunmin

CORPORATE SOURCE: Department of Internal Medicine, Konkuk University, Chungjoo, Chungbuk, Korea.

SOURCE: Life sciences, (2006 Jan 25) Vol. 78, No. 9, pp. 934-42.
Electronic Publication: 2005-09-22.
Journal code: 0375521. ISSN: 0024-3205.

PUB. COUNTRY: England: United Kingdom

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200602

ENTRY DATE: Entered STN: 24 Jan 2006
Last Updated on STN: 28 Feb 2006
Entered Medline: 27 Feb 2006

AB Extracts from Rhei Rhizoma extracts (RR) have been reported to attenuate metabolic disorders such as diabetic nephropathy, hypercholesterolemia and platelet aggregation. With this study we investigated the anti-diabetic action of 70% ethanol RR extract in streptozotocin-induced diabetic mice, and determined the action mechanism of active compounds of RR in vitro. In the diabetic mice, serum glucose levels at fasting and post-prandial states and glucose area under the curve at modified oral glucose tolerance tests were lowered without altering serum insulin levels, indicating that RR contained potential anti-diabetic agents. The fractions fractionated from RR extracts by XAD-4 column revealed that 60%, 80% and 100% methanol fractions enhanced insulin sensitivity and inhibited alpha-glucoamylase activity. The major compounds of these fractions were sennosides, rhein and rhaponticin. Rhaponticin and rhein enhanced insulin-stimulated glucose uptake in 3T3-L1 adipocytes. Rhaponticin increased adipocytes with a differentiating effect similar to pioglitazone, but rhein and sennoside B decreased triglyceride accumulation. Sennoside A and B inhibited alpha-glucoamylase activity as much as acarbose. In conclusion, a crude extract of RR improves glucose intolerance by enhancing insulin-stimulated glucose uptake and decreasing carbohydrate digestion via inhibiting alpha-glucoamylase activity. Rhein and rhaponticin are potential candidates for hypoglycemic agents.

L26 ANSWER 13 OF 18 MEDLINE on STN

ACCESSION NUMBER: 2002159587 MEDLINE

DOCUMENT NUMBER: PubMed ID: 11890888

TITLE: Addition of maltodextrins to the nonreducing-end of acarbose by reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase.

AUTHOR: Yoon Seung-Heon; Robyt John F

CORPORATE SOURCE: Laboratory of Carbohydrate Chemistry and Enzymology, Department of Biochemistry, Biophysics and Molecular Biology, Iowa State University, Ames, IA 50011, USA.

SOURCE: Carbohydrate research, (2002 Mar 15) Vol. 337, No. 6, pp. 509-16.
Journal code: 0043535. ISSN: 0008-6215.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200207

ENTRY DATE: Entered STN: 14 Mar 2002
Last Updated on STN: 23 Jul 2002
Entered Medline: 22 Jul 2002

AB New kinds of acarbose analogues were synthesized by the reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase (CGTase). Three major CGTase coupling products were separated and purified by Bio-Gel P2 gel-permeation chromatography. Digestion of the three products by beta-amylase and glucoamylase showed that they were composed of maltohexaose (G6), maltododecaose (G12), and maltooctadecaose (G18), respectively, attached to the nonreducing-end

of acarbose. ¹³C NMR of the glucoamylase product (D-glucopyranosyl-acarbose) showed that the D-glucose moiety was attached alpha- to the C-4-OH group of the nonreducing-end cyclohexene ring of acarbose, indicating that the maltodextrins were attached alpha-(1-->4) to the nonreducing-end cyclohexene of acarbose.

L26 ANSWER 14 OF 18 MEDLINE on STN
ACCESSION NUMBER: 90201033 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2108020
TITLE: Identification of carboxylic acid residues in glucoamylase G2 from *Aspergillus niger* that participate in catalysis and substrate binding.
AUTHOR: Svensson B; Clarke A J; Svendsen I; Moller H
CORPORATE SOURCE: Department of Chemistry, Carlsberg Laboratory, Copenhagen, Denmark.
SOURCE: European journal of biochemistry / FEBS, (1990 Feb 22) Vol. 188, No. 1, pp. 29-38.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199005
ENTRY DATE: Entered STN: 1 Jun 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 2 May 1990

AB Functionally important carboxyl groups in glucoamylase G2 from *Aspergillus niger* were identified using a differential labelling approach which involved modification of the acarbose-inhibited enzyme with 1-ethyl-3-(4-azonia-4,4-dimethylpentyl)carbodiimide (EAC) and inactivation by [3H]EAC following removal of acarbose. Subsequent sequence localization of the substituted acidic residues was facilitated by specific phenylthiohydantoins. The acid cluster Asp176, Glu179 and Glu180 reacted exclusively with [3H]EAC, while Asp112, Asp153, Glu259 and Glu389 had incorporated both [3H]EAC and EAC. It is conceivable that one or two of the [3H]EAC-labelled side chains act in catalysis while the other fully protected residue(s) participates in substrate binding probably together with the partially protected ones. Twelve carboxyl groups that reacted with EAC in the enzyme-acarbose complex were also identified. Asp176, Glu179 and Glu180 are all invariant in fungal glucoamylases. Glu180 was tentatively identified as a catalytic group on the basis of sequence alignments to catalytic regions in isomaltase and alpha-amylase. The partially radiolabelled Asp112 corresponds in Taka-amylase A to Tyr75 situated in a substrate binding loop at a distance from the site of cleavage. A possible correlation between carbodiimide modification of an essential carboxyl group and its role in the glucoamylase catalysis is discussed.

L26 ANSWER 15 OF 18 MEDLINE on STN
ACCESSION NUMBER: 88326243 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3415657
TITLE: Purification and characterization of a highly thermostable novel pullulanase from *Clostridium thermohydrosulfuricum*.
AUTHOR: Saha B C; Mathupala S P; Zeikus J G
CORPORATE SOURCE: Michigan Biotechnology Institute, Lansing 48909.
SOURCE: The Biochemical journal, (1988 Jun 1) Vol. 252, No. 2, pp. 343-8.
Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English

FILE SEGMENT: Priority Journals
ENTRY MONTH: 198809
ENTRY DATE: Entered STN: 8 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 28 Sep 1988

AB *Clostridium thermohydrosulfuricum* mutant Z 21-109 produced intracellular thermostable pullulanase and glucoamylase activities. The glucoamylase activity was inactivated by treating *C. thermohydrosulfuricum* cells with 10% (v/v) propan-1-ol at 85 degrees C in the presence of 5 mM-CaCl₂. Pullulanase activity was selectively solubilized from cells by treatment with detergent and lipase. The solubilized pullulanase was purified by treatment with streptomycin sulphate and (NH₄)₂SO₄ and by DEAE-Sephacel, octyl-Sepharose and pullulan-Sepharose chromatography. Pullulanase was purified 3511-fold and displayed homogeneity on SDS/polyacrylamide-gel electrophoresis. The pullulanase was a monomeric glycoprotein with an apparent Mr of about 136,500, and it displayed a pI of 5.9. The enzyme was enriched in both acidic and hydrophobic amino acids. The purified pullulanase was stable and optimally active at 90 degrees C. The optimum pH for activity and pH-stability ranges were 5.0-5.5 and 3.0-5.0 respectively. The enzyme was inhibited by cyclodextrins, EDTA and N-bromosuccinimide, but not by p-chloromercuribenzoate and acarbose. The pullulanase displayed a relative substrate specificity for hydrolysis of pullulan (100%) versus 75% for glycogen and 50% for soluble starch. The apparent Km, Vmax. and Kcat. values for enzyme activity on pullulan at 60 degrees C were 0.675 mg/ml, 122.5 mumol of reducing sugar formed/min per mg of protein and 16,240 min⁻¹ respectively. The novel properties of this extremely thermostable pullulanase are discussed in relation to other purified starch-debranching enzymes.

L26 ANSWER 16 OF 18 MEDLINE on STN
ACCESSION NUMBER: 87190439 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3106037
TITLE: Purification and characterization of extracellular alpha-amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678.
AUTHOR: De Mot R; Verachtert H
SOURCE: European journal of biochemistry / FEBS, (1987 May 4) Vol. 164, No. 3, pp. 643-54.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198706
ENTRY DATE: Entered STN: 3 Mar 1990
Last Updated on STN: 3 Mar 1990
Entered Medline: 25 Jun 1987

AB An alpha-amylase and a glucoamylase were purified to homogeneity from the culture fluid of beta-cyclodextrin-grown *Candida antarctica* CBS 6678 by protamine sulfate treatment, ammonium sulfate precipitation, gel filtration (Sephadex G-75 sf, Ultrogel AcA 54), DEAE-Sephacel chromatography, hydroxyapatite chromatography and affinity chromatography on acarbose--AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compositions. Their apparent relative molecular mass, sedimentation coefficient (S_{20,w}), isoelectric point, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2 and 57 degrees C, respectively, for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2 and 62 degrees C, respectively, for alpha-amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-molecular-mass substrates, including some raw starches. alpha-Amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase.

Trestatins were potent inhibitors of both alpha-amylase (K_i less than 1 μM) and glucoamylase (K_i less than 0.1 μM), being more effective than Bay e 4609 (K_i less than 10 μM). Glucoamylase was selectivity and strongly inhibited by acarbose (K_i less than 0.1 μM). Activity of the latter enzyme was also affected by 1-deoxynojirimycin (K_i less than 1 mM), maltitol and amino alcohols (K_i less than 10 mM). Unlike alpha-amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being non-identical with the active site.

L26 ANSWER 17 OF 18 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by
acarbose (BAY g-5421) affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L26 ANSWER 18 OF 18 MEDLINE on STN
ACCESSION NUMBER: 86129246 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3937490
TITLE: Purification and characterization of an
extracellular glucoamylase from the yeast *Candida*
tsukubaensis CBS 6389.
AUTHOR: De Mot R; Van Oudendijck E; Verachtert H
SOURCE: Antonie van Leeuwenhoek, (1985) Vol. 51, No. 3, pp. 275-87.
Journal code: 0372625. ISSN: 0003-6072.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198603
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 4 Mar 1986

AB The starch-degrading yeast *Candida tsukubaensis* CBS 6389 secreted amylase at high activity when grown in a medium containing soluble starch. The

extracellular alpha-amylase activity was very low. The major amylase component was purified by DEAE-Sephadex A-50 chromatography and Ultrogel AcA 44 gel filtration and characterized as a glucoamylase. The enzyme proved to be a glycoprotein with a molecular weight of 56 000. The glucoamylase had a temperature optimum at 55 degrees C and displayed highest activity in a pH range of 2.4-4.8. Acarbose strongly inhibited the purified glucoamylase. Debranching activity was present as demonstrated by the release of glucose from pullulan.

L28 ANSWER 1 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2006:65265 CAPLUS

DOCUMENT NUMBER: 144:226032

TITLE: Insulin sensitizing and α -glucoamylase
inhibitory action of sennosides, rhein and rhaponticin
in Rhei Rhizoma

AUTHOR(S): Choi, Soo Bong; Ko, Byoung Seob; Park, Seong Kyu;
Jang, Jin Sun; Park, Sunmin

CORPORATE SOURCE: Department of Internal Medicine, Konkuk University,
Chungjoo, Chungbuk, S. Korea

SOURCE: Life Sciences (2006), 78(9), 934-942
CODEN: LIFSAK; ISSN: 0024-3205

PUBLISHER: Elsevier B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Exts. from Rhei Rhizoma exts. (RR) have been reported to attenuate metabolic disorders such as diabetic nephropathy, hypercholesterolemia and platelet aggregation. This study was designed to investigate the anti-diabetic action of 70% ethanol RR extract in streptozotocin-induced diabetic mice, and determined the action mechanism of active compds. of RR in vitro. In the diabetic mice, serum glucose levels at fasting and post-prandial states and glucose area under the curve at modified oral glucose tolerance tests were lowered without altering serum insulin levels, indicating that RR contained potential anti-diabetic agents. The fractions fractionated from RR exts. by XAD-4 column revealed that 60%, 80% and 100% methanol fractions enhanced insulin sensitivity and inhibited α -glucoamylase activity. The major compds. of these fractions were sennosides, rhein and rhaponticin. Rhaponticin and rhein enhanced insulin-stimulated glucose uptake in 3T3-L1 adipocytes. Rhaponticin increased adipocytes with a differentiating effect similar to pioglitazone, but rhein and sennoside B decreased triglyceride accumulation. Sennoside A and B inhibited α -glucoamylase activity as much as acarbose. In conclusion, a crude extract of RR improves glucose intolerance by enhancing insulin-stimulated glucose uptake and decreasing carbohydrate digestion via inhibiting α -glucoamylase activity. Rhein and rhaponticin are potential candidates for hypoglycemic agents.

REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L28 ANSWER 2 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:67895 CAPLUS

DOCUMENT NUMBER: 136:288314

TITLE: Use of immobilised glucoamylase G2 for separation of
enantiomers

AUTHOR(S): Karlsson, A.; Arfwidsson, I.; Husovic, Z.; Svensson,
B.

CORPORATE SOURCE: Analytical R&D, AstraZeneca R and D Molndal, Moelndal,
43183, Swed.

SOURCE: Chromatographia (2001), 54(11/12), 717-723
CODEN: CHRGB7; ISSN: 0009-5893

PUBLISHER: Friedrich Vieweg & Sohn Verlagsgesellschaft mbH

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The aim of this study was to compare the isolated glucoamylase G2 domain as chiral selector with previously presented data obtained using immobilized native glucoamylase as chiral stationary phase. The glucoamylase G2 was isolated from a com. preparation of *Aspergillus niger* glucoamylase and immobilized onto silica particles. Enantioselectivity was tested for several amino alcs. including the β -receptor blocking agents metoprolol and alprenolol. Separation factors >2 were observed. Mobile phase pH was varied to optimize the enantioselective recognition and several different uncharged additives were included to

examine their effect on retention. For some of the tested solutes increased retention times were observed when increasing the content of uncharged modifier. Increased mobile phase concentration of several of the tested organic modifiers resulted in increased separation factors. The effect

of

column temperature was studied. Retention and enantioselectivity increased at higher temps. Addition to the mobile phase of acarbose, an inhibitor that binds to the catalytic site of G2 with picomolar affinity, resulted in total loss of enantioselectivity. Comparison of the present results with those obtained previously with the glucoamylase G1 form also containing a starch binding domain, shows that the catalytic domain is essential for chiral recognition of amino alcs.

REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L28 ANSWER 3 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1995:855392 CAPLUS

DOCUMENT NUMBER: 123:277181

TITLE: Functional role of *Aspergillus oryzae* glucoamylase C-terminal domain investigated using its cDNA

AUTHOR(S): Nagashima, Tadashi; Yamamoto, Yutaka; Kitamoto, Katushiko; Kumagai, Chieko

CORPORATE SOURCE: Technical service, Shin Nihon Chemical Co. Ltd., Aichi, 446, Japan

SOURCE: Journal of Fermentation and Bioengineering (1995), 80(3), 280-2

CODEN: JFBIEX; ISSN: 0922-338X

PUBLISHER: Society for Fermentation and Bioengineering, Japan

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The C-terminus domain of *Aspergillus oryzae* glucoamylase was analyzed by site-directed mutagenesis using glucoamylase cDNA. A mutant glucoamylase cDNA lacking the region corresponding to the C-terminal domain of the wild-type glucoamylase was constructed by inserting two stop codons in the gene for the wild-type glucoamylase. The wild-type and mutant glucoamylase cDNAs were expressed in *Saccharomyces cerevisiae* YPH 250, and then the produced wild-type and mutant glucoamylases were purified by acarbose affinity column chromatog. As compared to those of the wild-type glucoamylase, the K_m values of the mutant enzyme determined using maltose, maltotriose or maltopentaose as a substrate were similar, but that determined using soluble starch as a substrate was twofold

higher. The mutant glucoamylase showed a low rate of hydrolysis of raw cornstarch, although the wild-type glucoamylase showed a high rate of raw cornstarch hydrolysis. These results indicated that the C-terminal domain is important in the affinity of the enzyme to raw starch as reported for the glucoamylase of *Aspergillus awamori*.

L28 ANSWER 4 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1992:566290 CAPLUS

DOCUMENT NUMBER: 117:166290

TITLE: Rapid assay of glucoamylase using a fluorescence-labeled glucoamylase inhibitor, acarbose

AUTHOR(S): Hata, Yoji; Tanaka, Tatsuyuki; Suizu, Tetsuyoshi; Kawato, Akitsugu; Abe, Yasushisa; Imayasu, Satoshi; Ono, Kazuhisa; Oka, Satoru

CORPORATE SOURCE: Res. Inst. Gekkeikan, Gekkeikan Sake Co., Ltd., Kyoto, 612, Japan

SOURCE: Bioscience, Biotechnology, and Biochemistry (1992), 56(8), 1345-6

CODEN: BBBIEJ; ISSN: 0916-8451

DOCUMENT TYPE: Journal
LANGUAGE: English

AB Usually, glucoamylase activity was assayed by measuring the rate of release of glucose from soluble starch. However, in routine anal. the extract from rice-koji contains a large amount of glucose and oligosaccharides, and these saccharides affect the assay of the glucoamylase activity. Therefore, the koji-extract had to be dialyzed before the enzyme assay by the conventional methods. It has been reported however that the pseudooligosaccharides produced by *Streptomyces castaneglobisporus* inhibit fungal glucoamylases, and that affinity columns prepared with the immobilized glucoamylase inhibitors, the pseudooligosaccharides or acarbose, effectively adsorbed glucoamylase from unpasteurized sake. These observations suggested that the substantially high affinity of the inhibitor for glucoamylase (acarbose, $K_i = 0.5 \mu\text{M}$; maltose, $K_m = 1.1 \text{ mM}$) might be applicable to the assay of the enzyme. In this study, a rapid assay method for glucoamylases was developed using a fluorescence-labeled glucoamylase inhibitor. Acarbose and 2-aminopyridine were chosen as a glucoamylase inhibitor and a fluorescent reagent, resp. 2-Aminopyridine was coupled to acarbose. From 1 mg of acarbose, 300 μg of purified pyridylaminated (PA-) inhibitor was eluted as a single peak on Shim-pack CLC-ODS (M) (4.6 mm + 15 cm, Shimadzu), monitoring the glucoamylase-inhibitory activity and the fluorescent intensity. A protocol for glucoamylase assay using the fluorescence-labeled inhibitor is as follows. A glucoamylase solution to be assayed was reacted with the PA-inhibitor, and then the glucoamylase-bound PA-inhibitor was removed from the reaction mixture by an anion-exchange resin. Then, the glucoamylase activity in the sample was represented by the decrement in the fluorescent intensity (F_{dec}) that was given as the difference between the fluorescent intensity before (F_{int}) and after (F_{fin}) the elimination of the fluorescent affinity complex.

L28 ANSWER 5 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:566162 CAPLUS

DOCUMENT NUMBER: 109:166162

TITLE: Various molecular species in glucoamylase from *Aspergillus niger*

AUTHOR(S): Ono, Kazuhisa; Shintani, Koji; Shigeta, Seiko; Oka, Satoru

CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan

SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1689-98

CODEN: ABCHA6; ISSN: 0002-1369

DOCUMENT TYPE: Journal

LANGUAGE: English

AB An immobilized acarbose column selectively adsorbed most of glucoamylase components from a com. glucoamylase preps. The adsorbed enzyme was specifically eluted with maltose into a glucoamylase fraction free from α -amylase and α -glucosidase. The eluate was further fractionated into 6 subfractions by gel chromatog. and subsequent anion-exchange chromatog. Each of the enzyme subfractions liberated β -glucose as the sole product from soluble starch and maltooligosaccharides. Thus, all the enzymes are glucoamylases, though the enzymes were apparently discriminated from one another on the basis of mol. weight and(or) electrophoretic behavior. Furthermore, the enzyme subfractions were classified roughly into 3 groups on the structural resemblance implied by immunol. cross-reactivity among them.

L28 ANSWER 6 OF 8 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS

DOCUMENT NUMBER: 105:167504

TITLE: Purification of glucoamylase by
acarbose (BAY g-5421) affinity chromatography
AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.
CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA
SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3),
201-9
CODEN: BABIEC; ISSN: 0885-4513
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was
purified from com. available, impure enzyme preps. by affinity chromatog.
on acarbose (II) columns. Up to 2 mg I was bound
without leakage to a 1-mL affinity gel column possessing a
covalently linked II ligand (1 mg II/g wet gel), and the bound enzyme was
specifically released by irrigation of the column with a solution
of maltose. A complete cycle of purification was accomplished in .apprx.8 h.
Both I activities were recovered in >80% yield, free of α -amylase
activity and possessing specific activities comparable to those of preps.
obtained by time-consuming, multistep procedures involving several
ion-exchange and hydrophobic column fractionations. Thus, II
affinity chromatog. provides a general method for the rapid and efficient
purification of I, and appears to be ideally suited for scale-up for the com.
purification of these enzymes.

L28 ANSWER 7 OF 8 MEDLINE on STN
ACCESSION NUMBER: 2006040708 MEDLINE
DOCUMENT NUMBER: PubMed ID: 16182318
TITLE: Insulin sensitizing and alpha-glucoamylase inhibitory
action of sennosides, rheins and rhaponticin in Rhei
Rhizoma.
AUTHOR: Choi Soo Bong; Ko Byoung Seob; Park Seong Kyu; Jang Jin
Sun; Park Sunmin
CORPORATE SOURCE: Department of Internal Medicine, Konkuk University,
Chungjoo, Chungbuk, Korea.
SOURCE: Life sciences, (2006 Jan 25) Vol. 78, No. 9, pp. 934-42.
Electronic Publication: 2005-09-22.
Journal code: 0375521. ISSN: 0024-3205.
PUB. COUNTRY: England; United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200602
ENTRY DATE: Entered STN: 24 Jan 2006
Last Updated on STN: 28 Feb 2006
Entered Medline: 27 Feb 2006
AB Extracts from Rhei Rhizoma extracts (RR) have been reported to attenuate
metabolic disorders such as diabetic nephropathy, hypercholesterolemia and
platelet aggregation. With this study we investigated the anti-diabetic
action of 70% ethanol RR extract in streptozotocin-induced diabetic mice,
and determined the action mechanism of active compounds of RR in vitro.
In the diabetic mice, serum glucose levels at fasting and post-prandial
states and glucose area under the curve at modified oral glucose tolerance
tests were lowered without altering serum insulin levels, indicating that
RR contained potential anti-diabetic agents. The fractions fractionated
from RR extracts by XAD-4 column revealed that 60%, 80% and 100%
methanol fractions enhanced insulin sensitivity and inhibited alpha-
glucoamylase activity. The major compounds of these fractions
were sennosides, rhein and rhaponticin. Rhaponticin and rhein enhanced
insulin-stimulated glucose uptake in 3T3-L1 adipocytes. Rhaponticin
increased adipocytes with a differentiating effect similar to
pioglitazone, but rhein and sennoside B decreased triglyceride
accumulation. Sennoside A and B inhibited alpha-glucoamylase
activity as much as acarbose. In conclusion, a crude extract of
RR improves glucose intolerance by enhancing insulin-stimulated glucose

uptake and decreasing carbohydrate digestion via inhibiting alpha-glucoamylase activity. Rhein and rhaponticin are potential candidates for hypoglycemic agents.

L28 ANSWER 8 OF 8 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose
(BAY g-5421) affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L29 ANSWER 10 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:419287 CAPLUS

DOCUMENT NUMBER: 109:19287

TITLE: Purification and characterization of a highly thermostable novel pullulanase from *Clostridium thermohydrosulfuricum*

AUTHOR(S): Saha, Badal C.; Mathupala, Saroj P.; Zeikus, J. Gregory

CORPORATE SOURCE: Michigan Biotechnol. Inst., Lansing, MI, 48909, USA

SOURCE: Biochemical Journal (1988), 252(2), 343-8

CODEN: BIJOAK; ISSN: 0306-3275

DOCUMENT TYPE: Journal

LANGUAGE: English

AB *C. thermohydrosulfuricum* Mutant Z 21-109 produced intracellular thermostable pullulanase and glucoamylase activities. The glucoamylase activity was inactivated by treating *C. thermohydrosulfuricum* cells with 10% propan-1-ol at 85° in the presence of 5 mM CaCl₂. Pullulanase activity was selectively solubilized from cells by treatment with detergent and lipase. The solubilized pullulanase was purified by treatment with streptomycin sulfate and (NH₄)₂SO₄ and by DEAE-Sephacel, octyl-Sepharose, and pullulan-Sepharose chromatog. Pullulanase was purified 3511-fold and displayed homogeneity on SDS-PAGE. The pullulanase was a monomeric glycoprotein with an apparent mol. weight of .apprx.136,500, and it displayed a pI of 5.9. The enzyme was enriched in both acidic and hydrophobic amino acids. The purified pullulanase was stable and optimally active at 90°. The optimum pH for activity and pH-stability ranges were 5.0-5.5 and 3.0-5.0, resp. The enzyme was inhibited by cyclodextrins, EDTA, and N-bromosuccinimide, but not by p-chloromercuribenzoate and acarbose. Pullulanase displayed a relative substrate specificity for hydrolysis of pullulan (100%) vs. 75% for glycogen and 50% for soluble starch. The apparent Km, Vmax, and catalytic rate constant values for enzyme activity on pullulan at 60° were 0.675 mg/mL, 122.5 µmol of reducing sugar formed/min/mg of protein, and 16,240 min⁻¹ resp. The novel properties of this extremely thermostable pullulanase are discussed in relation to other purified starch-debranching enzymes.

L29 ANSWER 11 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:209924 CAPLUS

DOCUMENT NUMBER: 106:209924

TITLE: Purification and characterization of extracellular α-amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678

AUTHOR(S): De Mot, Rene; Verachtert, Hubert

CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Heverlee, B-3030, Belg.

SOURCE: European Journal of Biochemistry (1987), 164(3), 643-54

CODEN: EJBCAI; ISSN: 0014-2956

DOCUMENT TYPE: Journal

LANGUAGE: English

AB An α-amylase and a glucoamylase were purified to homogeneity from the culture fluid of β-cyclodextrin-grown *C. antarctica* CBS 6678 by protamine sulfate treatment, (NH₄)₂SO₄ precipitation, gel filtration (Sephadex G-75 sf, Ultrogel Aca 54), DEAE-Sephacel chromatog., hydroxyapatite chromatog., and affinity chromatog. on acarbose-AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compns. Their apparent relative mol. mass, sedimentation coefficient (s_{20,w}), pI, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2°, and 57°, resp., for

glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2° and 62°, resp., for α -amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-mol.-mass substrates, including some raw starches. The α -amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both α -amylase ($K_i < 1 \mu\text{M}$) and glucoamylase ($K_i < 0.1 \mu\text{M}$), being more effective than Bay e 4609 ($K_i < 10 \mu\text{M}$). Glucoamylase was selectively and strongly inhibited by acarbose ($K_i < 0.1 \mu\text{M}$). Activity of the latter enzyme was also affected by 1-deoxynojirimycin ($K_i < 1 \text{ mM}$), maltitol, and amino alcs. ($K_i < 10 \text{ mM}$). Unlike α -amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being nonidentical with the active site.

L29 ANSWER 12 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS

DOCUMENT NUMBER: 105:167504

TITLE: Purification of glucoamylase by acarbose (BAY g-5421) affinity chromatography

AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.

CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA

SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3), 201-9

CODEN: BABIEC; ISSN: 0885-4513

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was purified from com. available, impure enzyme preps. by affinity chromatog. on acarbose (II) columns. Up to 2 mg I was bound without leakage to a 1-mL affinity gel column possessing a covalently linked II ligand (1 mg II/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in .apprx.8 h. Both I activities were recovered in >80% yield, free of α -amylase activity and possessing specific activities comparable to those of preps. obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, II affinity chromatog. provides a general method for the rapid and efficient purification of I, and appears to be ideally suited for scale-up for the com. purification of these enzymes.

L29 ANSWER 13 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:125516 CAPLUS

DOCUMENT NUMBER: 104:125516

TITLE: Purification and characterization of an extracellular glucoamylase from the yeast *Candida tsukubaensis* CBS 6389

AUTHOR(S): De Mot, R.; Van Oudendijck, E.; Verachtert, H.

CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Louvain, B-3030, Belg.

SOURCE: Antonie van Leeuwenhoek (1985), 51(3), 275-87

CODEN: ALJMAO; ISSN: 0003-6072

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The starch-degrading yeast *C. tsukubaensis* CBS 6389 secreted amylase with high activity when grown in a medium containing soluble starch. The extracellular α -amylase activity was very low. The major amylase component was purified by DEAE-Sephadex A-50 chromatog. and Ultrogel AcA 44 gel filtration and characterized as a glucoamylase. The enzyme was a glycoprotein with a mol. weight of 56,000. The glucoamylase had a temperature optimum of 55°, and its highest activity was at pH 2.4-4.8. Acarbose strongly inhibited the purified glucoamylase. Debranching activity was present, as

demonstrated by the release of glucose from pullulan.

L29 ANSWER 14 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:30774 CAPLUS

DOCUMENT NUMBER: 104:30774

TITLE: Purification and characterization of extracellular amylolytic enzymes from the yeast *Filobasidium capsuligenum*

AUTHOR(S): De Mot, Rene; Verachtert, Hubert

CORPORATE SOURCE: Lab. Ind. Microbiol. Biochem., Univ. Leuven, Louvain, B-3030, Belg.

SOURCE: Applied and Environmental Microbiology (1985), 50(6), 1474-82

CODEN: AEMIDF; ISSN: 0099-2240

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The extracellular amylolytic system of *Filobasidium capsuligenum* consisted of an α -amylase (EC 3.2.1.1) (I) and 2 forms of glucoamylase (EC 3.2.1.3) (II). The enzymes were purified by (NH₄)₂SO₄ fractionation, repeated ion-exchange chromatog. (DEAE-Sephadex A-50), and gel filtration (Sephadex G-25, Sephadex G-100). I had an optimum pH of 5.6 and an optimum temperature of 50°, but was rapidly inactivated at higher temperature. The mol. weight was estimated by SDS-polyacrylamide gel electrophoresis to be 64,000. An acarbose concentration of 20 μ g/mL was required for 50% inhibition of I. Both II enzymes were glycoproteins of identical mol. weight (60,000) and produced only glucose by exohydrolysis. The debranching activity of the II enzymes was evidenced with substrates containing α -1,6 linkages. The pH optima were 5.0-5.6 for II-I and 4.8-5.3 for II-II. II-I had a higher optimum temperature (55%) than II-II (50%) and was also more resistant to thermal inactivation. Only low acarbose concns. (<0.1 μ g/mL) were required to reduce the activity of II-I and II-II by 50%.

L29 ANSWER 15 OF 20 MEDLINE on STN

ACCESSION NUMBER: 2002159587 MEDLINE

DOCUMENT NUMBER: PubMed ID: 11890888

TITLE: Addition of maltodextrins to the nonreducing-end of acarbose by reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase.

AUTHOR: Yoon Seung-Heon; Robyt John F

CORPORATE SOURCE: Laboratory of Carbohydrate Chemistry and Enzymology, Department of Biochemistry, Biophysics and Molecular Biology, Iowa State University, Ames, IA 50011, USA.

SOURCE: Carbohydrate research, (2002 Mar 15) Vol. 337, No. 6, pp. 509-16.

Journal code: 0043535. ISSN: 0008-6215.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200207

ENTRY DATE: Entered STN: 14 Mar 2002

Last Updated on STN: 23 Jul 2002

Entered Medline: 22 Jul 2002

AB New kinds of acarbose analogues were synthesized by the reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase (CGTase). Three major CGTase coupling products were separated and purified by Bio-Gel P2 gel-permeation chromatography. Digestion of the three products by beta-amylase and glucoamylase showed that they were composed of maltohexaose (G6), maltododecaose (G12), and maltooctadecaose (G18), respectively, attached to the nonreducing-end of acarbose. ¹³C NMR of the glucoamylase product (D-glucopyranosyl-acarbose) showed

that the D-glucose moiety was attached alpha- to the C-4-OH group of the nonreducing-end cyclohexene ring of acarbose, indicating that the maltodextrins were attached alpha-(1-->4) to the nonreducing-end cyclohexene of acarbose.

L29 ANSWER 16 OF 20 MEDLINE on STN
ACCESSION NUMBER: 90201033 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2108020
TITLE: Identification of carboxylic acid residues in glucoamylase G2 from *Aspergillus niger* that participate in catalysis and substrate binding.
AUTHOR: Svensson B; Clarke A J; Svendsen I; Moller H
CORPORATE SOURCE: Department of Chemistry, Carlsberg Laboratory, Copenhagen, Denmark.
SOURCE: European journal of biochemistry / FEBS, (1990 Feb 22) Vol. 188, No. 1, pp. 29-38.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199005
ENTRY DATE: Entered STN: 1 Jun 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 2 May 1990

AB Functionally important carboxyl groups in glucoamylase G2 from *Aspergillus niger* were identified using a differential labelling approach which involved modification of the acarbose-inhibited enzyme with 1-ethyl-3-(4-azonia-4,4-dimethylpentyl)carbodiimide (EAC) and inactivation by [3H]EAC following removal of acarbose. Subsequent sequence localization of the substituted acidic residues was facilitated by specific phenylthiohydantoins. The acid cluster Asp176, Glu179 and Glu180 reacted exclusively with [3H]EAC, while Asp112, Asp153, Glu259 and Glu389 had incorporated both [3H]EAC and EAC. It is conceivable that one or two of the [3H]EAC-labelled side chains act in catalysis while the other fully protected residue(s) participates in substrate binding probably together with the partially protected ones. Twelve carboxyl groups that reacted with EAC in the enzyme-acarbose complex were also identified. Asp176, Glu179 and Glu180 are all invariant in fungal glucoamylases. Glu180 was tentatively identified as a catalytic group on the basis of sequence alignments to catalytic regions in isomaltase and alpha-amylase. The partially radiolabelled Asp112 corresponds in Taka-amylase A to Tyr75 situated in a substrate binding loop at a distance from the site of cleavage. A possible correlation between carbodiimide modification of an essential carboxyl group and its role in the glucoamylase catalysis is discussed.

L29 ANSWER 17 OF 20 MEDLINE on STN
ACCESSION NUMBER: 88326243 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3415657
TITLE: Purification and characterization of a highly thermostable novel pullulanase from *Clostridium thermohydrosulfuricum*.
AUTHOR: Saha B C; Mathupala S P; Zeikus J G
CORPORATE SOURCE: Michigan Biotechnology Institute, Lansing 48909.
SOURCE: The Biochemical journal, (1988 Jun 1) Vol. 252, No. 2, pp. 343-8.
Journal code: 2984726R. ISSN: 0264-6021.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198809
ENTRY DATE: Entered STN: 8 Mar 1990

Last Updated on STN: 3 Feb 1997

Entered Medline: 28 Sep 1988

- AB *Clostridium thermohydrosulfuricum* mutant Z 21-109 produced intracellular thermostable pullulanase and glucoamylase activities. The glucoamylase activity was inactivated by treating *C. thermohydrosulfuricum* cells with 10% (v/v) propan-1-ol at 85 degrees C in the presence of 5 mM-CaCl₂. Pullulanase activity was selectively solubilized from cells by treatment with detergent and lipase. The solubilized pullulanase was purified by treatment with streptomycin sulphate and (NH₄)₂SO₄ and by DEAE-Sephacel, octyl-Sepharose and pullulan-Sepharose chromatography. Pullulanase was purified 3511-fold and displayed homogeneity on SDS/polyacrylamide-gel electrophoresis. The pullulanase was a monomeric glycoprotein with an apparent Mr of about 136,500, and it displayed a pI of 5.9. The enzyme was enriched in both acidic and hydrophobic amino acids. The purified pullulanase was stable and optimally active at 90 degrees C. The optimum pH for activity and pH-stability ranges were 5.0-5.5 and 3.0-5.0 respectively. The enzyme was inhibited by cyclodextrins, EDTA and N-bromosuccinimide, but not by p-chloromercuribenzoate and acarbose. The pullulanase displayed a relative substrate specificity for hydrolysis of pullulan (100%) versus 75% for glycogen and 50% for soluble starch. The apparent Km, Vmax. and Kcat. values for enzyme activity on pullulan at 60 degrees C were 0.675 mg/ml, 122.5 mumol of reducing sugar formed/min per mg of protein and 16,240 min⁻¹ respectively. The novel properties of this extremely thermostable pullulanase are discussed in relation to other purified starch-debranching enzymes.

L29 ANSWER 18 OF 20 MEDLINE on STN

ACCESSION NUMBER: 87190439 MEDLINE

DOCUMENT NUMBER: PubMed ID: 3106037

TITLE: Purification and characterization of extracellular alpha-amylase and glucoamylase from the yeast *Candida antarctica* CBS 6678.

AUTHOR: De Mot R; Verachtert H

SOURCE: European journal of biochemistry / FEBS, (1987 May 4) Vol. 164, No. 3, pp. 643-54.

Journal code: 0107600. ISSN: 0014-2956.

PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198706

ENTRY DATE: Entered STN: 3 Mar 1990

Last Updated on STN: 3 Mar 1990

Entered Medline: 25 Jun 1987

- AB An alpha-amylase and a glucoamylase were purified to homogeneity from the culture fluid of beta-cyclodextrin-grown *Candida antarctica* CBS 6678 by protamine sulfate treatment, ammonium sulfate precipitation, gel filtration (Sephadex G-75 sf, Ultrogel AcA 54), DEAE-Sephacel chromatography, hydroxyapatite chromatography and affinity chromatography on acarbose--AH-Sepharose 4B. Both enzymes were monomeric glycoproteins with fairly different amino acid compositions. Their apparent relative molecular mass, sedimentation coefficient (S_{20,w}), isoelectric point, absorption coefficient (280 nm), pH and temperature optima were estimated as 48,500, 4.7 S, 10.1, 1.74 cm² mg⁻¹, 4.2 and 57 degrees C, respectively, for glucoamylase and as 50,000, 4.9 S, 10.3, 1.53 cm² mg⁻¹, 4.2 and 62 degrees C, respectively, for alpha-amylase. Kinetic analyses indicated that both enzymes preferentially hydrolyzed high-molecular-mass substrates, including some raw starches. alpha-Amylase was active on cyclodextrins, whereas debranching activity was demonstrated for glucoamylase. Trestatins were potent inhibitors of both alpha-amylase (K_i less than 1 microM) and glucoamylase (K_i less than 0.1 microM), being more

effective than Bay e 4609 (K_i less than 10 microM). Glucoamylase was selectivity and strongly inhibited by acarbose (K_i less than 0.1 microM). Activity of the latter enzyme was also affected by 1-deoxynojirimycin (K_i less than 1 mM), maltitol and amino alcohols (K_i less than 10 mM). Unlike alpha-amylase, glucoamylase adsorbed strongly onto raw starch, the adsorption site being non-identical with the active site.

L29 ANSWER 19 OF 20 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose
(BAY g-5421) affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol.
8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these enzymes.

L29 ANSWER 20 OF 20 MEDLINE on STN
ACCESSION NUMBER: 86129246 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3937490
TITLE: Purification and characterization of an extracellular glucoamylase from the yeast *Candida tsukubaensis* CBS 6389.
AUTHOR: De Mot R; Van Oudendijck E; Verachtert H
SOURCE: *Antonie van Leeuwenhoek*, (1985) Vol. 51, No. 3, pp. 275-87.
Journal code: 0372625. ISSN: 0003-6072.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198603
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Feb 1997
Entered Medline: 4 Mar 1986

AB The starch-degrading yeast *Candida tsukubaensis* CBS 6389 secreted amylase at high activity when grown in a medium containing soluble starch. The extracellular alpha-amylase activity was very low. The major amylase component was purified by DEAE-Sephadex A-50 chromatography and Ultrogel AcA 44 gel filtration and characterized as a glucoamylase

. The enzyme proved to be a glycoprotein with a molecular weight of 56 000. The glucoamylase had a temperature optimum at 55 degrees C and displayed highest activity in a pH range of 2.4-4.8. Acarbose strongly inhibited the purified glucoamylase. Debranching activity was present as demonstrated by the release of glucose from pullulan.

L29 ANSWER 1 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|------|----------|-----------------|----------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |

PRIORITY APPLN. INFO.: TW 2003-92133913 A 20031202
AB A purification process for manufacturing a high pure acarbose relates to a
process
for preparing high pure acarbose from acarbose-containing fermentation broth.
The
acarbose was purified through steps of alc. precipitation, a strongly acidic
cation exchanger chromatog. and an immobilized enzyme affinity
chromatog. Acarbose is generally applied in treating diabetes.

L29 ANSWER 2 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:1068953 CAPLUS
DOCUMENT NUMBER: 142:273282
TITLE: A New Method for Screening α -Glucosidase
Inhibitors and Application to Marine Microorganisms
AUTHOR(S): Chen, Haimin; Yan, Xiaojun; Lin, Wei; Zheng, Li;
Zhang, Weiwei
CORPORATE SOURCE: Institute of Oceanology, Chinese Academy of Sciences,
Qingdao, Peop. Rep. China
SOURCE: Pharmaceutical Biology (Lisse, Netherlands) (2004),
42(6), 416-421
CODEN: PHBIFC; ISSN: 1388-0209
PUBLISHER: Taylor & Francis The Netherlands
DOCUMENT TYPE: Journal
LANGUAGE: English
AB A new enzyme assay method for screening α -glucosidase inhibitors
with rapidity and simplicity was developed. The enzyme-substituted
 α -glucosidases for this assay was glucoamylase. Samples were
spotted or developed on the silica gel plate. The agar solution containing
substrate was poured on the plate, and paper impregnated with enzyme was
layered on the agar. After incubation, an inhibitory circle would appear
around the inhibitor. By using this method, more than 200 strains of
marine microorganisms were screened. Among them, three active strains
were found to secrete inhibitors in the culture medium.
REFERENCE COUNT: 9 THERE ARE 9 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L29 ANSWER 3 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2003:356465 CAPLUS
DOCUMENT NUMBER: 138:334026
TITLE: High throughput isolation of biological compounds
INVENTOR(S): Frisner, Henrik; Christensen, Lars Lehmann Hylling
PATENT ASSIGNEE(S): Novozymes A/S, Den.
SOURCE: PCT Int. Appl., 37 pp.
CODEN: PIXXD2

DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|--|----------|-----------------|----------|
| WO 2003037914 | A2 | 20030508 | WO 2002-DK717 | 20021029 |
| WO 2003037914 | A3 | 20040325 | | |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| CA 2465543 | AA | 20030508 | CA 2002-2465543 | 20021029 |
| AU 2002340776 | A1 | 20030512 | AU 2002-340776 | 20021029 |
| EP 1442048 | A2 | 20040804 | EP 2002-774472 | 20021029 |
| R: | AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, SK | | | |

PRIORITY APPLN. INFO.: DK 2001-1598 A 20011030
WO 2002-DK717 W 20021029

AB A method for high throughput micro-purification of a library of tag-free biol. compds. is claimed wherein a population of discrete liquid samples comprising the library of tag-free biol. compds. is contacted with a solid chromatog. material comprising a functional group selected from ion exchange materials, hydrophobic materials, affinity materials and hydrophobic charge induction materials so as to retain the tag-free biol. compds. of the library, isolating the chromatog. material retaining the biol. compds., releasing the biol. compds. of the library from the solid chromatog. material and collecting the released biol. compds. of the library to produce a population of samples comprising the isolated biol. compound

L29 ANSWER 4 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:249053 CAPLUS

DOCUMENT NUMBER: 136:401960

TITLE: Addition of maltodextrins to the nonreducing-end of acarbose by reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase

AUTHOR(S): Yoon, Seung-Heon; Robyt, John F.

CORPORATE SOURCE: Laboratory of Carbohydrate Chemistry and Enzymology, Department of Biochemistry, Biophysics and Molecular Biology, Iowa State University, Ames, IA, 50011, USA

SOURCE: Carbohydrate Research (2002), 337(6), 509-516

CODEN: CRBRAT; ISSN: 0008-6215

PUBLISHER: Elsevier Science Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

OTHER SOURCE(S): CASREACT 136:401960

AB New kinds of acarbose analogs were synthesized by the reaction of acarbose with cyclomaltohexaose and cyclomaltodextrin glucanyltransferase (CGTase). Three major CGTase coupling products were separated and purified by Bio-Gel P2 gel-permeation chromatog. Digestion of the three products by beta-amylase and glucoamylase showed that they were composed of maltohexaose (G6), maltododecaose (G12), and maltooctadecaose (G18), resp., attached to the nonreducing-end of acarbose. ¹³C NMR of the glucoamylase product (D-glucopyranosyl-acarbose) showed that the D-glucose moiety was

attached α - to the C-4-OH group of the nonreducing-end cyclohexene ring of acarbose, indicating that the maltodextrins were attached α -(1 \rightarrow 4) to the nonreducing-end cyclohexene of acarbose.

REFERENCE COUNT: 26 THERE ARE 26 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L29 ANSWER 5 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:67895 CAPLUS

DOCUMENT NUMBER: 136:288314

TITLE: Use of immobilised glucoamylase G2 for separation of enantiomers

AUTHOR(S): Karlsson, A.; Arfwidsson, I.; Husovic, Z.; Svensson, B.

CORPORATE SOURCE: Analytical R&D, AstraZeneca R and D Molndal, Moelndal, 43183, Swed.

SOURCE: Chromatographia (2001), 54(11/12), 717-723

CODEN: CHRGB7; ISSN: 0009-5893

PUBLISHER: Friedrich Vieweg & Sohn Verlagsgesellschaft mbH

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The aim of this study was to compare the isolated glucoamylase G2 domain as chiral selector with previously presented data obtained using immobilized native glucoamylase as chiral stationary phase. The glucoamylase G2 was isolated from a com. preparation of *Aspergillus niger* glucoamylase and immobilized onto silica particles. Enantioselectivity was tested for several amino alcs. including the β -receptor blocking agents metoprolol and alprenolol. Separation factors >2 were observed. Mobile phase pH was varied to optimize the enantioselective recognition and several different uncharged additives were included to examine their effect on retention. For some of the tested solutes increased retention times were observed when increasing the content of uncharged modifier. Increased mobile phase concentration of several of the tested organic modifiers resulted in increased separation factors. The effect

of

column temperature was studied. Retention and enantioselectivity increased at higher temps. Addition to the mobile phase of acarbose, an inhibitor that binds to the catalytic site of G2 with picomolar affinity, resulted in total loss of enantioselectivity. Comparison of the present results with those obtained previously with the glucoamylase G1 form also containing a starch binding domain, shows that the catalytic domain is essential for chiral recognition of amino alcs.

REFERENCE COUNT: 28 THERE ARE 28 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L29 ANSWER 6 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1995:855392 CAPLUS

DOCUMENT NUMBER: 123:277181

TITLE: Functional role of *Aspergillus oryzae* glucoamylase C-terminal domain investigated using its cDNA

AUTHOR(S): Nagashima, Tadashi; Yamamoto, Yutaka; Kitamoto, Katushiko; Kumagai, Chieko

CORPORATE SOURCE: Technical service, Shin Nihon Chemical Co. Ltd., Aichi, 446, Japan

SOURCE: Journal of Fermentation and Bioengineering (1995), 80(3), 280-2

CODEN: JFBIEX; ISSN: 0922-338X

PUBLISHER: Society for Fermentation and Bioengineering, Japan

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The C-terminus domain of *Aspergillus oryzae* glucoamylase was analyzed by site-directed mutagenesis using glucoamylase cDNA. A mutant glucoamylase cDNA lacking the region corresponding to the C-terminal domain of the wild-type glucoamylase was

constructed by inserting two stop codons in the gene for the wild-type glucoamylase. The wild-type and mutant glucoamylase cDNAs were expressed in *Saccharomyces cerevisiae* YPH 250, and then the produced wild-type and mutant glucoamylases were purified by acarbose affinity column chromatog. As compared to those of the wild-type glucoamylase, the K_m values of the mutant enzyme determined using maltose, maltotriose or maltopentaose as a substrate were similar, but that determined using soluble starch as a substrate was twofold higher. The mutant glucoamylase showed a low rate of hydrolysis of raw cornstarch, although the wild-type glucoamylase showed a high rate of raw cornstarch hydrolysis. These results indicated that the C-terminal domain is important in the affinity of the enzyme to raw starch as reported for the glucoamylase of *Aspergillus awamori*.

L29 ANSWER 7 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1994:100047 CAPLUS
DOCUMENT NUMBER: 120:100047
TITLE: Purification and characterization of a pullulan-hydrolyzing glucoamylase from *Sclerotium rolfsii*
AUTHOR(S): Kelkar, Hemant S.; Deshpande, Mukund V.
CORPORATE SOURCE: Pune, India
SOURCE: Starch/Staerke (1993), 45(10), 361-8
CODEN: STARDD; ISSN: 0038-9056
DOCUMENT TYPE: Journal
LANGUAGE: English

AB The pullulan-hydrolyzing enzyme from culture filtrates of *Sclerotium rolfsii* grown on soluble starch as a carbon source has been purified by ultrafiltration (Amicon, PM-10), ion-exchange chromatog. (DEAE-Cellulose DE-52) and gel filtration chromatog. (Bio-Gel P-150). The enzyme moved as a single band in non-denaturing polyacrylamide gel electrophoresis carried out at pH 2.9 and 7.5. The relative mol. mass of the enzyme was estimated to be 64,000 D by SDS-PAGE and 66,070 D by gel filtration on Bio-Gel P150. The enzyme hydrolyzed pullulan optimally at 50°C between pH 4.0-4.5, whereas, soluble starch was optimally hydrolyzed at a pH of between 4.0-4.5 and at 65°C. The Michaelis constant (K_m) for pullulan was 5.13 mg·mL⁻¹ (V_{max} 1.0 U·mg⁻¹) and for soluble starch, it was 0.6 mg·mL⁻¹ (V_{max} 8.33 U·mg⁻¹). The enzyme was observed to be a glycoprotein (12-13% carbohydrate by weight) and had a strong affinity for Con A. The enzyme hydrolyzed α -D-glucans in an exo-manner, which resulted in the release of glucose as the sole product of hydrolysis. Acarbose, a maltotetraose analog, was found to be a potent inhibitor of both pullulan and starch hydrolysis (100% inhibition at 0.06 μ M). The enzyme has been characterized as a glucoamylase (1,4- α -D-glucan glucohydrolase, EC 3.2.1.3) showing a significant action on pullulan.

L29 ANSWER 8 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:566162 CAPLUS
DOCUMENT NUMBER: 109:166162
TITLE: Various molecular species in glucoamylase from *Aspergillus niger*
AUTHOR(S): Ono, Kazuhisa; Shintani, Koji; Shigeta, Seiko; Oka, Satoru
CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan
SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1689-98
CODEN: ABCHA6; ISSN: 0002-1369
DOCUMENT TYPE: Journal
LANGUAGE: English

AB An immobilized acarbose column selectively adsorbed most of

glucoamylase components from a com. glucoamylase prepns. The adsorbed enzyme was specifically eluted with maltose into a glucoamylase fraction free from α -amylase and α -glucosidase. The eluate was further fractionated into 6 subfractions by gel chromatog. and subsequent anion-exchange chromatog. Each of the enzyme subfractions liberated β -glucose as the sole product from soluble starch and maltooligosaccharides. Thus, all the enzymes are glucoamylases, though the enzymes were apparently discriminated from one another on the basis of mol. weight and(or) electrophoretic behavior. Furthermore, the enzyme subfractions were classified roughly into 3 groups on the structural resemblance implied by immunol. cross-reactivity among them.

L29 ANSWER 9 OF 20 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:524834 CAPLUS

DOCUMENT NUMBER: 109:124834

TITLE: Effective purification of glucoamylase in koji, a solid culture of *Aspergillus oryzae* on steamed rice, by affinity chromatography using an immobilized acarbose (BAY g-5421)

AUTHOR(S): Ono, Kazuhisa; Shigeta, Seiko; Oka, Satoru

CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan

SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1707-14

CODEN: ABCHA6; ISSN: 0002-1369

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Glucoamylase (GA) was purified from koji, a solid culture of *A. oryzae* on steamed rice, by extraction with 1% NaCl solution, precipitation with EtOH, and acarbose affinity chromatog. The purified enzyme was homogeneous on gel filtration, PAGE and SDS-PAGE, ultracentrifugation, and IEF. The enzyme released β -glucose as a sole product from soluble starch and maltooligosaccharides. The other common and inherent features of GAs were also confirmed in the GA from *A. oryzae*. The enzyme was a glycoprotein containing .apprx.4.8% glucosamine and 7.8% neutral saccharides.

L30 ANSWER 1 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:999873 CAPLUS

DOCUMENT NUMBER: 143:272742

TITLE: Identification of impurities in acarbose by using an integrated liquid chromatography-nuclear magnetic resonance and liquid chromatography-mass spectrometry approach

AUTHOR(S): Novak, Predrag; Cindric, Mario; Tepes, Predrag; Dragojevic, Snjezana; Ilijas, Marina; Mihaljevic, Kreso

CORPORATE SOURCE: PLIVA Research Institute, Zagreb, Croatia

SOURCE: Journal of Separation Science (2005), 28(13), 1442-1447

CODEN: JSSCCJ; ISSN: 1615-9306

PUBLISHER: Wiley-VCH Verlag GmbH & Co. KGaA

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The usefulness of applying an integrated LC-NMR and LC-MS approach to acarbose bulk drug impurity profiling is demonstrated. LC-MS and LC-NMR methodologies were employed for the online separation and structural elucidation of a final drug product. Combining data provided by the stop-flow LC-NMR and LC-MS expts. made it possible to identify the main components present in the acarbose sample. Spectral anal. revealed that A and B were known impurities while C was an unknown compound. LC-MS and LC-NMR analyses revealed that C was a pentasaccharide differing from the acarbose in number and nature of sugar subunits in the mol. It was subsequently isolated and its structure was confirmed by the off-line 1- and 2-D NMR expts., and atom assignment was made.

REFERENCE COUNT: 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L30 ANSWER 2 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:440618 CAPLUS

DOCUMENT NUMBER: 141:49832

TITLE: Quantification of Acarbose in Human Plasma by Liquid Chromatography-Electrospray Tandem Mass Spectrometry

AUTHOR(S): Raut, B. B.; Kolte, B. L.; Deo, A. A.; Bagool, M. A.; Shinde, D. B.

CORPORATE SOURCE: Wockhardt Research Centre, Maharashtra, India

SOURCE: Journal of Liquid Chromatography & Related Technologies (2004), 27(11), 1759-1768

CODEN: JLCTFC; ISSN: 1082-6076

PUBLISHER: Marcel Dekker, Inc.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The method for the determination of acarbose in human plasma is described, using

HPLC separation with tandem mass spectrometric detection. Samples were prepared

using solid phase extraction and separated on a Zorbax SB C18 column with a mobile

phase consisting of H₂O, MeCN, and trifluoroacetic acid. Detection was performed by a TSQ quantum mass spectrometer in the selected reaction monitoring (SRM) mode using electrospray ionization (ESI). The method has a chromatog. elution time of 3 min and was linear within the range of 100-1000 ng/mL. The intra- and inter-run accuracy and precision, calculated from quality control (QC) samples, was <11%.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L30 ANSWER 3 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:191789 CAPLUS

DOCUMENT NUMBER: 141:49901
TITLE: Combined use of liquid chromatography
-nuclear magnetic resonance spectroscopy and liquid
chromatography-mass spectrometry for the
characterization of an acarbose degradation
product
AUTHOR(S): Novak, Predrag; Tepes, Predrag; Cindric, Mario;
Ilijas, Marina; Dragojevic, Snjezana; Mihaljevic,
Kreso
CORPORATE SOURCE: PLIVA Research Institute Ltd., Zagreb, HR-10000,
Croatia
SOURCE: Journal of Chromatography, A (2004), 1033(2), 299-303
CODEN: JCRAEY; ISSN: 0021-9673
PUBLISHER: Elsevier Science B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Directly coupled LC-MS and LC-NMR were applied to identify and
structurally characterize an acarbose degradation product A in acidic media.
A comparative anal. of the stop-flow LC-NMR (1H and TOCSY) and LC-MS data
provided evidence that A is structurally related to acarbose, differing
from the parent compound in a number of subunits present in the mol. Spectral
anal. revealed that A was the α -glucosidase inhibitor amylostatin
XG. Complementary information obtained from the two methods led to the
structural elucidation of A which was later corroborated by high-resolution
NMR spectroscopy of the isolated mol.
REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L30 ANSWER 4 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:331546 CAPLUS
DOCUMENT NUMBER: 137:72514
TITLE: High performance liquid chromatography of
acarbose and its metabolite on porous
graphitic carbon column
AUTHOR(S): Daali, Youssef; Cherkaoui, Samir; Cahours, Xavier;
Varesio, Emmanuel; Veuthey, Jean-Luc
CORPORATE SOURCE: Laboratory of Pharmaceutical Analytical Chemistry,
University of Geneva, Geneva, 1211/4, Switz.
SOURCE: Journal of Separation Science (2002), 25(5/6), 280-284
CODEN: JSSCCJ; ISSN: 1615-9306
PUBLISHER: Wiley-VCH Verlag GmbH
DOCUMENT TYPE: Journal
LANGUAGE: English
AB This paper describes the development of an anal. method for the determination
of
two pseudo-oligosaccharides, acarbose and its main metabolite. The anal.
was carried out by liquid chromatog. on a porous graphitic C stationary
phase. The separation mechanism of these compds. is dominated by
charge-induced interactions between the polar analyte and the polarizable
surface of graphitic C. Several chromatog. parameters were studied,
including the nature and percentage of the organic solvent and acid modifier
as well as column temperature. The best conditions were achieved with a mobile
phase containing HO₂CCF₃ 0.1% and MeCN 8%. Under these conditions, the
simultaneous resolution of acarbose and its metabolite and of their anomers
was achieved, highlighting the potential of this stationary phase for
challenging sepn. Also, three detection methods are described and
compared in terms of sensitivity. The evaporative light scattering
detector was not sensitive enough for acarbose and metabolite determination in
biol. fluids. Mass spectrometry allowed a significant improvement in
sensitivity but was not sufficient to permit the anal. of selected compds.
in such complex matrixes. However, this detector confirmed the presence
of acarbose and its metabolite as well as the anomeric separation. Finally,
fluorescence detection is very sensitive after derivatization with
2-aminobenzamide and enabled the determination of selected compds. at a low
concentration

level.

REFERENCE COUNT: 17 THERE ARE 17 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L30 ANSWER 5 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1999:62094 CAPLUS

DOCUMENT NUMBER: 130:242375

TITLE: Development and validation of liquid chromatography and capillary electrophoresis methods for acarbose determination in pharmaceutical tablets

AUTHOR(S): Cherkaoui, S.; Daali, Y.; Christen, P.; Veuthey, J.-L.

CORPORATE SOURCE: Laboratory of Pharmaceutical Analytical Chemistry, University of Geneva, Geneva, 1211, Switz.

SOURCE: Journal of Pharmaceutical and Biomedical Analysis (1998), 18(4,5), 729-735

CODEN: JPBADA; ISSN: 0731-7085

PUBLISHER: Elsevier Science B.V.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Liquid chromatog. and capillary zone electrophoresis, resp. coupled to an evaporative light scattering detector and a UV detector were developed for the anal. of acarbose without any derivatization procedure. The electrophoretic separation of acarbose anomers was achieved through the manipulation of the working temperature Both methods were validated and showed good validation data in terms of precision, accuracy and linearity. The validated methods were successfully applied to the dosage of acarbose in com. available Glucobay tablets.

REFERENCE COUNT: 19 THERE ARE 19 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L30 ANSWER 6 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1994:293511 CAPLUS

DOCUMENT NUMBER: 120:293511

TITLE: Gas chromatographic-mass spectrometric determination of ¹³C-glucose level for evaluating the effect of α -glucosidase inhibitor acarbose on the digestion of [U-¹³C]starch in rat

AUTHOR(S): Goromaru, Tsuyoshi; Matsuki, Kazuhiro; Matsuki, Yoko

CORPORATE SOURCE: Fac. Pharm. Pharm. Sci., Fukuyama Univ., Fukuyama, 729-02, Japan

SOURCE: Biological & Pharmaceutical Bulletin (1994), 17(1), 156-9

CODEN: BPBLEO; ISSN: 0918-6158

DOCUMENT TYPE: Journal

LANGUAGE: English

AB [U-¹³C]starch was administered orally to rats with or without acarbose. After the addition of [2H₃]-D-glucose as the internal standard, the plasma samples were treated successively for defatting, deproteinizing and desalting. Glucose was converted to sorbitol by reduction with sodium borohydride. The cyclic butylboronate of sorbitol was injected into a gas chromatograph-mass spectrometer, and the concentration of labeled glucose was measured by selected monitoring of the quasi-mol. ion. The plasma concentration of labeled glucose was decreased significantly by the addition of acarbose. The effect of acarbose on the digestion of starch was clearly confirmed using [U-¹³C]starch.

L30 ANSWER 7 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1988:524834 CAPLUS

DOCUMENT NUMBER: 109:124834

TITLE: Effective purification of glucoamylase in koji, a solid culture of *Aspergillus oryzae* on steamed rice,

by affinity chromatography using an immobilized acarbose (BAY g-5421)
 AUTHOR(S): Ono, Kazuhisa; Shigeta, Seiko; Oka, Satoru
 CORPORATE SOURCE: Fac. Eng., Hiroshima Univ., Higashi-Hiroshima, 724, Japan
 SOURCE: Agricultural and Biological Chemistry (1988), 52(7), 1707-14
 CODEN: ABCHA6; ISSN: 0002-1369
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Glucoamylase (GA) was purified from koji, a solid culture of *A. oryzae* on steamed rice, by extraction with 1% NaCl solution, precipitation with EtOH, and acarbose affinity chromatog. The purified enzyme was homogeneous on gel filtration, PAGE and SDS-PAGE, ultracentrifugation, and IEF. The enzyme released β -glucose as a sole product from soluble starch and maltooligosaccharides. The other common and inherent features of GAs were also confirmed in the GA from *A. oryzae*. The enzyme was a glycoprotein containing .apprx.4.8% glucosamine and 7.8% neutral saccharides.

L30 ANSWER 8 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1987:497062 CAPLUS
 DOCUMENT NUMBER: 107:97062
 TITLE: Purification of acarbose via chromatography on weakly acidic cation exchange resins

INVENTOR(S): Rauenbusch, Erich
 PATENT ASSIGNEE(S): Bayer A.-G. , Fed. Rep. Ger.
 SOURCE: Ger. Offen., 7 pp.
 CODEN: GWXXBX

DOCUMENT TYPE: Patent
 LANGUAGE: German
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|----------|
| DE 3543999 | A1 | 19870619 | DE 1985-3543999 | 19851213 |
| EP 226121 | A2 | 19870624 | EP 1986-116773 | 19861202 |
| EP 226121 | A3 | 19890322 | | |
| EP 226121 | B1 | 19920122 | | |
| R: AT, BE, CH, DE, ES, FR, GB, IT, LI, NL, SE | | | | |
| AT 71951 | E | 19920215 | AT 1986-116773 | 19861202 |
| ES 2038591 | T3 | 19930801 | ES 1986-116773 | 19861202 |
| JP 62155288 | A2 | 19870710 | JP 1986-292667 | 19861210 |
| JP 2502551 | B2 | 19960529 | | |
| US 4904769 | A | 19900227 | US 1986-940713 | 19861211 |
| CA 1288768 | A1 | 19910910 | CA 1986-525014 | 19861211 |
| DK 8605986 | A | 19870614 | DK 1986-5986 | 19861212 |
| DK 164870 | B | 19920831 | | |
| DK 164870 | C | 19930111 | | |
| HU 43083 | A2 | 19870928 | HU 1986-5203 | 19861212 |
| HU 196219 | B | 19881028 | | |
| CN 86108259 | A | 19870729 | CN 1986-108259 | 19861213 |
| CN 1013866 | B | 19910911 | | |
| JP 08245683 | A2 | 19960924 | JP 1996-34402 | 19960129 |
| JP 2628853 | B2 | 19970709 | | |

PRIORITY APPLN. INFO.: DE 1985-3543999 A 19851213
 EP 1986-116773 A 19861202

AB Acarbose (I) containing <10% impurity (excluding H₂O) is prepared by passing a 1-20% solution of I at pH 4-7 through a column packed with a dextran-, agarose-, or cellulose-based carboxylic acid ion exchanger optionally containing a polyamide. A solution of 9.2 g impure I in .apprx.40 mL H₂O adjusted to pH 4.7 was added to a 2.6 + 34 cm column containing

CM-Sephadex C 25 previously treated with a NaOAc buffer and flushed with H₂O. I was eluted with H₂O at 100 mL/h to give 5.87 g of 93% pure I.

L30 ANSWER 9 OF 14 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 1986:567504 CAPLUS
DOCUMENT NUMBER: 105:167504
TITLE: Purification of glucoamylase by acarbose
(BAY g-5421) affinity chromatography
AUTHOR(S): Ono, Kazuhisa; Smith, Eric E.
CORPORATE SOURCE: Sch. Med., Univ. Miami, Miami, FL, 33101, USA
SOURCE: Biotechnology and Applied Biochemistry (1986), 8(2-3),
201-9
CODEN: BABIEC; ISSN: 0885-4513
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Glucoamylase (I) of *Aspergillus niger* and *Rhizopus* species was purified from com. available, impure enzyme prepns. by affinity chromatog. on acarbose (II) columns. Up to 2 mg I was bound without leakage to a 1-mL affinity gel column possessing a covalently linked II ligand (1 mg II/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in .apprx.8 h. Both I activities were recovered in >80% yield, free of α -amylase activity and possessing specific activities comparable to those of prepns. obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, II affinity chromatog. provides a general method for the rapid and efficient purification of I, and appears to be ideally suited for scale-up for the com. purification of these enzymes.

L30 ANSWER 10 OF 14 MEDLINE on STN

ACCESSION NUMBER: 2005488560 MEDLINE
DOCUMENT NUMBER: PubMed ID: 16158985
TITLE: Identification of impurities in acarbose by using
an integrated liquid chromatography-nuclear
magnetic resonance and liquid chromatography-mass
spectrometry approach.
AUTHOR: Novak Predrag; Cindric Mario; Tepes Predrag; Dragojevic
Snjezana; Ilijas Marina; Mihaljevic Kreso
CORPORATE SOURCE: PLIVA Research Institute, Prilaz baruna Filipovica 29,
Zagreb, Croatia.. predrag.novak@pliva.hr
SOURCE: Journal of separation science, (2005 Aug) Vol. 28, No. 13,
pp. 1442-7.
Journal code: 101088554. ISSN: 1615-9306.
PUB. COUNTRY: Germany: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200608
ENTRY DATE: Entered STN: 15 Sep 2005
Last Updated on STN: 17 Aug 2006
Entered Medline: 16 Aug 2006

AB The usefulness of applying an integrated LC-NMR and LC-MS approach to acarbose bulk drug impurity profiling is demonstrated. LC-MS and LC-NMR methodologies were employed for the online separation and structural elucidation of a final drug product. Combining data provided by the stop-flow LC-NMR and LC-MS experiments made it possible to identify the main components present in the acarbose sample. Spectral analysis revealed that A and B were known impurities while C was an unknown compound. LC-MS and LC-NMR analyses revealed that C was a pentasaccharide differing from the acarbose in number and nature of sugar subunits in the molecule. It was subsequently isolated and its structure was confirmed by the offline 1- and 2-D NMR experiments, and atom assignment was made.

L30 ANSWER 11 OF 14 MEDLINE on STN

ACCESSION NUMBER: 2004192762 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 15088751
 TITLE: Combined use of liquid chromatography-nuclear magnetic resonance spectroscopy and liquid chromatography-mass spectrometry for the characterization of an acarbose degradation product.
 AUTHOR: Novak Predrag; Tepes Predrag; Cindric Mario; Ilijas Marina; Dragojevic Snjezana; Mihaljevic Kreso
 CORPORATE SOURCE: PLIVA Research Institute Ltd., Prilaz baruna Filipovica 29, HR-10000 Zagreb, Croatia.. predrag.novak@pliva.hr
 SOURCE: Journal of chromatography. A, (2004 Apr 16) Vol. 1033, No. 2, pp. 299-303.
 Journal code: 9318488. ISSN: 0021-9673.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 200411
 ENTRY DATE: Entered STN: 20 Apr 2004
 Last Updated on STN: 3 Nov 2004
 Entered Medline: 2 Nov 2004
 AB Directly coupled LC-MS and LC-NMR were applied to identify and structurally characterize an acarbose degradation product A in acidic media. A comparative analysis of the stop-flow LC-NMR (1H and TOCSY) and LC-MS data provided evidence that A is structurally related to acarbose, differing from the parent compound in a number of subunits present in the molecule. Spectral analysis revealed that A was the alpha-glucosidase inhibitor amylostatin XG. Complementary information obtained from the two methods led to the structural elucidation of A which was later corroborated by high-resolution NMR spectroscopy of the isolated molecule.

L30 ANSWER 12 OF 14 MEDLINE on STN
 ACCESSION NUMBER: 1999116703 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 9919975
 TITLE: Development and validation of liquid chromatography and capillary electrophoresis methods for acarbose determination in pharmaceutical tablets.
 AUTHOR: Cherkaoui S; Daali Y; Christen P; Veuthey J L
 CORPORATE SOURCE: Laboratory of Pharmaceutical Analytical Chemistry, University of Geneva, Switzerland.
 SOURCE: Journal of pharmaceutical and biomedical analysis, (1998 Dec) Vol. 18, No. 4-5, pp. 729-35.
 Journal code: 8309336. ISSN: 0731-7085.
 PUB. COUNTRY: ENGLAND: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199904
 ENTRY DATE: Entered STN: 26 Apr 1999
 Last Updated on STN: 3 Mar 2000
 Entered Medline: 13 Apr 1999
 AB Liquid chromatography and capillary zone electrophoresis, respectively coupled to an evaporative light scattering detector and a UV detector have been developed for the analysis of acarbose without any derivatization procedure. The electrophoretic separation of acarbose anomers was achieved through the manipulation of the working temperature. Both methods were validated and showed good validation data in terms of precision, accuracy and linearity. The validated methods were successfully applied to the dosage of acarbose in commercially available Glucobay tablets.

L30 ANSWER 13 OF 14 MEDLINE on STN
 ACCESSION NUMBER: 94198815 MEDLINE

DOCUMENT NUMBER: PubMed ID: 8148808
TITLE: Gas chromatographic-mass spectrometric determination of ¹³C-glucose level for evaluating the effect of alpha-glucosidase inhibitor acarbose on the digestion of [U-¹³C] starch in rat.
AUTHOR: Goromaru T; Matsuki K; Matsuki Y
CORPORATE SOURCE: Faculty of Pharmacy and Pharmaceutical Sciences, Fukuyama University, Japan.
SOURCE: Biological & pharmaceutical bulletin, (1994 Jan) Vol. 17, No. 1, pp. 156-9.
Journal code: 9311984. ISSN: 0918-6158.
PUB. COUNTRY: Japan
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199405
ENTRY DATE: Entered STN: 23 May 1994
Last Updated on STN: 3 Mar 2000
Entered Medline: 11 May 1994

AB The effect of acarbose on the digestion of starch was examined by a stable isotope tracer technique. [U-¹³C]-Starch was administered orally to rats with or without acarbose. After the addition of [2H₃]-D-glucose as the internal standard, the plasma samples were treated successively for defatting, deproteinizing and desalting. Glucose was converted to sorbitol by reduction with sodium borohydride. The cyclic butylboronate of sorbitol was injected into a gas chromatograph-mass spectrometer, and the concentration of labeled glucose was measured by selected monitoring of the quasi-molecular ion. The plasma concentration of labeled glucose was decreased significantly by the addition of acarbose. The effect of acarbose on the digestion of starch was clearly confirmed using [U-¹³C]starch.

L30 ANSWER 14 OF 14 MEDLINE on STN
ACCESSION NUMBER: 86296199 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3091050
TITLE: Purification of glucoamylase by acarbose (BAY g-5421) affinity chromatography.
AUTHOR: Ono K; Smith E E
CONTRACT NUMBER: DE-03118 (NIDCR)
SOURCE: Biotechnology and applied biochemistry, (1986 Apr-Jun) Vol. 8, No. 2-3, pp. 201-9.
Journal code: 8609465. ISSN: 0885-4513.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198610
ENTRY DATE: Entered STN: 21 Mar 1990
Last Updated on STN: 3 Mar 2000
Entered Medline: 23 Oct 1986

AB *Aspergillus niger* and *Rhizopus* sp. glucoamylases were purified on an affinity chromatography column from commercially available, impure enzyme preparations. Up to 2 mg of glucoamylase protein was bound without leakage to a 1-ml affinity gel column (0.7 X 2.5 cm) possessing a covalently linked acarbose ligand (1 mg acarbose/g wet gel), and the bound enzyme was specifically released by irrigation of the column with a solution of maltose. A complete cycle of purification was accomplished in about 8 h. Glucoamylases were recovered, in more than 80% yield, free of alpha-amylase activity and possessing specific activities comparable to those of preparations obtained by time-consuming, multistep procedures involving several ion-exchange and hydrophobic column fractionations. Thus, acarbose affinity chromatography provides a general method for the rapid and efficient purification of the glucoamylases, and seems to be ideally suited for scale-up for the commercial purification of these

enzymes.

L32 ANSWER 1 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purification process for manufacturing a high pure acarbose relates to a process for preparing high pure acarbose from acarbose-containing fermentation broth. The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L32 ANSWER 2 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:928233 CAPLUS
DOCUMENT NUMBER: 138:3755
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Hung.
SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S. Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|--|----------|-----------------|-------------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | |
| RW: | GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | US 2001-924271 | A2 20010807 |

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing

the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of a sodium chloride solution and a salt solution; precipitating the acarbose with a solvent; and recovering the precipitated acarbose.

L32 ANSWER 3 OF 3 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:123021 CAPLUS
DOCUMENT NUMBER: 136:182542
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.
SOURCE: PCT Int. Appl., 24 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---|------|----------|-----------------|------------|
| WO 2002012256 | A1 | 20020214 | WO 2001-US24729 | 20010807 |
| W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM | | | | |
| RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG | | | | |
| AU 2001084741 | A5 | 20020218 | AU 2001-84741 | 20010807 |
| EP 1309601 | A1 | 20030514 | EP 2001-963821 | 20010807 |
| R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR | | | | |
| PRIORITY APPLN. INFO.: | | | US 2000-223492P | P 20000807 |
| | | | WO 2001-US24729 | W 20010807 |

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of hydrochloric acid and the weak acid; precipitating the acarbose with a solvent; and separating the acarbose crystals.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L33 ANSWER 1 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2005:474833 CAPLUS
DOCUMENT NUMBER: 143:6386
TITLE: Purification process for manufacturing a high purity
acarbose
INVENTOR(S): Lin, Chung-Liang; Huang, Tung-Li; Chen, Jeen-Kuan; Wu,
Chi-Sheng
PATENT ASSIGNEE(S): Taiwan
SOURCE: U.S. Pat. Appl. Publ., 10 pp.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|------------------|------------|
| US 2005118686 | A1 | 20050602 | US 2004-790069 | 20040302 |
| JP 2005160463 | A2 | 20050623 | JP 2004-1337 | 20040106 |
| PRIORITY APPLN. INFO.: | | | TW 2003-92133913 | A 20031202 |

AB A purification process for manufacturing a high pure acarbose relates to a process for preparing high pure acarbose from acarbose-containing fermentation broth. The acarbose was purified through steps of alc. precipitation, a strongly acidic cation exchanger chromatog. and an immobilized enzyme affinity chromatog. Acarbose is generally applied in treating diabetes.

L33 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2004:924641 CAPLUS
DOCUMENT NUMBER: 142:155800
TITLE: Process for preparing α -glycosidase inhibitor
derived from powdery silkworm
INVENTOR(S): Ahn, Mi Yeong; Kim, Eun Seon; Kim, Gwang Won; Kim, Ik
Su; Kim, Jin Won; Lee, Hui Sam; Lee, Yong Gi; Ryu,
Gang Seon; Seo, Su Won
PATENT ASSIGNEE(S): Republic of Korea Management : Rural Development
Administration, S. Korea; Samsung Life Public Welfare
Foundation Samsung Medical Center
SOURCE: Repub. Korean Kongkae Taeho Kongbo, No pp. given
CODEN: KRXXA7
DOCUMENT TYPE: Patent
LANGUAGE: Korean
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|------------------------|------|----------|-----------------|----------|
| KR 2002071334 | A | 20020912 | KR 2001-11449 | 20010306 |
| PRIORITY APPLN. INFO.: | | | KR 2001-11449 | 20010306 |

AB Provided is a process for preparing α -glycosidase inhibitor derived from powdery silkworm, which has the same decreasing effect of blood glucose level as acarbose which is a conventional therapeutic agent for diabetes. The process for preparing α -glycosidase inhibitor comprises the steps of: freeze-drying and powdering silkworm then adding 50% ethanol to extract it; performing cation exchange column chromatog. with the extract then followed by eluting it with 0.5N of NH_4OH and concentrating it; dissolving the concentrate in water then followed by performing anion exchange column chromatog. eluting with water and concentrating it; dissolving the concentrate in water then followed by cation exchange column chromatog., eluting with water and 0.5N of NH_4OH to obtain 5 fractions obtain one fraction resp.; and performing anion exchange column chromatog.

with the second fraction among the 6 fractions to obtain pure 1-deoxynojirimycin fraction.

L33 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:928233 CAPLUS
DOCUMENT NUMBER: 138:3755
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Hung.
SOURCE: U.S. Pat. Appl. Publ., 7 pp., Cont.-in-part of U. S. Ser. No. 924,271.
CODEN: USXXCO
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|------|----------|-----------------|----------|
| US 2002183262 | A1 | 20021205 | US 2002-60831 | 20020130 |
| US 2002111320 | A1 | 20020815 | US 2001-924271 | 20010807 |
| WO 2003014135 | A1 | 20030220 | WO 2002-US2705 | 20020130 |

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM

RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2000-223492P P 20000807
US 2001-924271 A2 20010807

AB The present invention relates to a novel process for the preparation of acarbose. Said process comprises the steps of: acidifying a fermentation broth containing an acarbose; removing particulates from the fermentation broth; adsorbing the acarbose on a cation-exchanger in the presence of an anion of a weak acid; eluting the acarbose from the cation-exchanger with at least one of a sodium chloride solution and a salt solution; precipitating the acarbose with a solvent; and recovering the precipitated acarbose.

L33 ANSWER 4 OF 4 CAPLUS COPYRIGHT 2006 ACS on STN

ACCESSION NUMBER: 2002:123021 CAPLUS
DOCUMENT NUMBER: 136:182542
TITLE: Method for purification of acarbose
INVENTOR(S): Keri, Vilmos; Deak, Lajos
PATENT ASSIGNEE(S): Biogal Gyogyszergyar Rt., Hung.; Teva Pharmaceuticals USA, Inc.
SOURCE: PCT Int. Appl., 24 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

| PATENT NO. | KIND | DATE | APPLICATION NO. | DATE |
|---------------|---|----------|-----------------|----------|
| WO 2002012256 | A1 | 20020214 | WO 2001-US24729 | 20010807 |
| W: | AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, | | | |

LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT,
RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US,
UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF,
BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

AU 2001084741 A5 20020218 AU 2001-84741 20010807

EP 1309601 A1 20030514 EP 2001-963821 20010807

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

PRIORITY APPLN. INFO.: US 2000-223492P P 20000807

WO 2001-US24729 W 20010807

AB The present invention relates to a novel process for the preparation of
acarbose. Said process comprises the steps of: acidifying a fermentation broth
containing an acarbose; removing particulates from the fermentation broth;
adsorbing
the acarbose on a cation-exchanger in the presence of an anion of a weak
acid; eluting the acarbose from the cation-exchanger with at least one of
hydrochloric acid and the weak acid; precipitating the acarbose with a solvent;
and separating the acarbose crystals.

REFERENCE COUNT: 3 THERE ARE 3 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT